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Trying 31060000009999...Open

DIALOG INFORMATION SERVICES
PLEASE LOGON:
 ******** HHHHHHHH SSSSSSSS? ### Status: Signing onto Dialog ********
ENTER PASSWORD:
 ******** HHHHHHHH SSSSSSSS?  ********
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### Status: Login successfulDialog level 05.16.01D

Last logoff:  21feb07 14:03:15
Logon file405 28feb07 16:01:52
            *** ANNOUNCEMENTS ***
                      ***
NEW FILES RELEASED
***Engineering Index Backfile (File 988)
***EMCare (File 45)
***Trademarkscan - South Korea (File 655)

RESUMED UPDATING
***File 141, Reader's Guide Abstracts
                    ***
RELOADS COMPLETED
***Files 340, 341 & 942, CLAIMS/U.S. Patents - 2006 reload now online
***Files 173 & 973, Adis Clinical Trials Insight
                                ***
DATABASES REMOVED

Chemical Structure Searching now available in Prous Science Drug
Data Report (F452), Prous Science Drugs of the Future (F453),
IMS R&D Focus (F445/955), Pharmaprojects (F128/928), Beilstein
Facts (F390), Derwent Chemistry Resource (F355) and Index Chemicus
(File 302).
                    ***
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 >>>a specific database by entering HELP NEWS <file number>.<<<
* * *
SYSTEM:HOME
Cost is in DialUnits
Menu System II: D2 version 1.8.0 term=ASCII
                      *** DIALOG HOMEBASE(SM) Main Menu ***

 Information:
  1.  Announcements (new files, reloads, etc.)
  2.  Database, Rates, & Command Descriptions
  3.  Help in Choosing Databases for Your Topic
  4.  Customer Services (telephone assistance, training, seminars, etc.)
  5.  Product Descriptions

 Connections:
  6.  DIALOG(R) Document Delivery
  7.  Data Star(R)

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      /H = Help            /L = Logoff          /NOMENU = Command Mode


Enter an option number to view information or to connect to an online
 service.  Enter a BEGIN command plus a file number to search a database
(e.g., B1 for ERIC).
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Terminal set to DLINK
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 Information:
  1.  Announcements (new files, reloads, etc.)
  2.  Database, Rates, & Command Descriptions
  3.  Help in Choosing Databases for Your Topic
  4.  Customer Services (telephone assistance, training, seminars, etc.)
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      /H = Help            /L = Logoff          /NOMENU = Command Mode


Enter an option number to view information or to connect to an online
 service.  Enter a BEGIN command plus a file number to search a database
(e.g., B1 for ERIC).
? b biosci

>>>          44 is unauthorized
>>>          76 is unauthorized
>>>2 of the specified files are not available
       28feb07 16:01:57 User276653 Session D86.1
            $0.00    0.245 DialUnits FileHomeBase
     $0.00  Estimated cost FileHomeBase
     $0.02  TELNET
     $0.02  Estimated cost this search
     $0.02  Estimated total session cost   0.245 DialUnits


SYSTEM:OS  - DIALOG OneSearch
  File   5:Biosis Previews(R) 1969-2007/Feb W3
         (c) 2007 The Thomson Corporation
  File  24:CSA Life Sciences Abstracts 1966-2007/Nov
         (c) 2007 CSA.
  File  28:Oceanic Abstracts 1966-2007/Nov
         (c) 2007 CSA.
  File  34:SciSearch(R) Cited Ref Sci 1990-2007/Feb W3
         (c) 2007 The Thomson Corp
  File  35:Dissertation Abs Online 1861-2007/Feb
         (c) 2007 ProQuest Info&Learning
  File  40:Enviroline(R) 1975-2007/Jan
         (c) 2007 Congressional Information Service
  File  41:Pollution Abstracts 1966-2007/Nov
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        (c) 2007 CSA.
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        (c) 2007 Elsevier B.V.
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        (c) 2007 CAB International
  File  65:Inside Conferences 1993-2007/Feb 28
        (c) 2007 BLDSC all rts. reserv.
  File  71:ELSEVIER BIOBASE 1994-2007/Feb W4
        (c) 2007 Elsevier B.V.
  File  73:EMBASE 1974-2007/Feb 28
        (c) 2007 Elsevier B.V.
  File  91:MANTIS(TM) 1880-2006/Jan
        2001 (c) Action Potential
  File  94:JICST-EPlus 1985-2007/Mar W1
        (c)2007 Japan Science and Tech Corp(JST)
*File  94: UD200609W2 is the last update for 2006. UD200701W1 is the
first update for 2007. The file is complete and up to date.
  File  98:General Sci Abs 1984-2007/Feb
        (c) 2007 The HW Wilson Co.
  File 110:WasteInfo 1974-2002/Jul
        (c)  2002 AEA Techn Env.
*File 110: This file is closed (no updates)
  File 135:NewsRx Weekly Reports 1995-2007/Feb W3
        (c) 2007 NewsRx
  File 136:BioEngineering Abstracts 1966-2007/Nov
        (c) 2007 CSA.
  File 143:Biol. & Agric. Index 1983-2007/Feb
        (c) 2007 The HW Wilson Co
  File 144:Pascal 1973-2007/Feb W3
        (c) 2007 INIST/CNRS
  File 155:MEDLINE(R) 1950-2007/Feb 26
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  File 164:Allied & Complementary Medicine 1984-2007/Mar
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  File 172:EMBASE Alert 2007/Feb 28
        (c) 2007 Elsevier B.V.
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        (c) 2007 The Thomson Corp.
  File 357:Derwent Biotech Res. _1982-2007/Feb W3
        (c) 2007 The Thomson Corp.
  File 369:New Scientist 1994-2007/Nov W1
        (c) 2007 Reed Business Information Ltd.
  File 370:Science 1996-1999/Jul W3
        (c) 1999 AAAS
*File 370: This file is closed (no updates). Use File 47 for more current
information.
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        (c) 2006 Beilstein GmbH
  File 434:SciSearch(R) Cited Ref Sci 1974-1989/Dec
        (c) 2006 The Thomson Corp
  File 467:ExtraMED(tm) 2000/Dec
        (c) 2001 Informania Ltd.

     Set  Items  Description
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? s aspen and SP1
          22940  ASPEN
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          46954  SP1
      S1      33  ASPEN AND SP1
? t s1/9,k/1-33

 1/9,K/1     (Item 1 from file: 5)
DIALOG(R)File   5:Biosis Previews(R)
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19241949   BIOSIS NO.: 200600587344
 Aspen   SP1 , an exceptional thermal, protease and detergent-resistant
  self-assembled nano-particle
AUTHOR: Wang Wang-Xia; Dgany Or; Wolf Sharon Grayer; Levy Ilan; Algom
  Rachel; Pouny Yehonathan; Wolf Amnon; Marton Ira; Altman Arie; Shoseyov
  Oded (Reprint)
AUTHOR ADDRESS: Hebrew Univ Jerusalem, Fac Agr Food and Environm Qual Sci,
  Robert H Smith Inst Plant Sci and Genet Agriculture, POB 12, IL-76100
  Rehovot, Israel**Israel
AUTHOR E-MAIL ADDRESS: shoseyov@agri.huji.ac.il
JOURNAL: Biotechnology and Bioengineering  95 (1): p161-168 SEP 5 2006 2006
ISSN: 0006-3592
DOCUMENT TYPE: Article
RECORD TYPE: Abstract
LANGUAGE: English

ABSTRACT: Stable protein 1 ( SP1 ) is a homo-oligomeric protein isolated
  from  aspen  (Populus tremula  aspen ) plants which forms a ring-shape
  dodecameric particle with a central cavity. The oligomeric form of  SP1
  is an exceptionally stable structure that is resistant to proteases
  (e.g., trypsin, V8, and proteinase K), high temperatures, organic
  solvents, and high levels of ionic detergent. Analytical
  ultra-centrifugation, chemical cross-linking, matrix-assisted
  laser-desorption time-of-flight mass spectrometry (MALDI-TOF-MS), and
  transmission electron microscopy were used to further characterize the
  SP1  dodecamer. Introduction of a single cysteine at the N-terminus of
  SP1  enabled the formation of disulfide bridges within the  SP1
  dodecamer, concurrent with increased melting point. A six-histidine tag
  was introduced at the N-terminus of  SP1  to generate 6HSP1, and the
  Delta NSP1 mutant was generated by a deletion of amino acids 2-6 at the
  N-terminus. Both 6HSP1 and ANSP1 maintained their ability to assemble a
  stable dodecamer. Remarkably, these  SP1  homo-dodecamers were able to
  re-assemble into stable hetero-dodecamers following co-electro-elution
  from SDS-PAGE. The exceptional stability of the  SP1 -nano ring and its
  ability to self-assemble hetero-complexes paves the way to further
  research in utilizing this unique protein in nano-biotechnology. (c) 2006
  Wiley Periodicals, Inc.

REGISTRY NUMBERS: 9002-07-7: trypsin; 3374-22-9: cysteine; 4998-57-6:
    histidine; 39450-01-6: proteinase K; 16734-12-6: disulfide; 9001-92-7:
    protease
ENZYME COMMISSION NUMBER: EC 3.4.21.4: trypsin; EC 3.4.21.64: proteinase K
DESCRIPTORS:
  MAJOR CONCEPTS: Enzymology--Biochemistry and Molecular Biophysics;
    Biomaterials
  BIOSYSTEMATIC NAMES: Salicaceae--Dicotyledones, Angiospermae,
    Spermatophyta, Plantae
  ORGANISMS: Populus tremula { aspen } (Salicaceae)
  COMMON TAXONOMIC TERMS: Angiosperms; Dicots; Plants; Spermatophytes;
```

Vascular Plants
CHEMICALS & BIOCHEMICALS: trypsin; cysteine; histidine; proteinase K; disulfide; protease; V8; stable protein 1 { **SP1** }
METHODS & EQUIPMENT: self-assembled nano-particle--drug delivery device
MISCELLANEOUS TERMS: temperature range; self-assembly; ionic detergent
CONCEPT CODES:
10064 Biochemistry studies - Proteins, peptides and amino acids
10511 Biophysics - Bioengineering
10802 Enzymes - General and comparative studies: coenzymes
51518 Plant physiology - Enzymes
BIOSYSTEMATIC CODES:
26695 Salicaceae

Aspen SP1 , **an exceptional thermal, protease and detergent-resistant self-assembled nano-particle**
ABSTRACT: Stable protein 1 (**SP1**) is a homo-oligomeric protein isolated from **aspen** (Populus tremula **aspen**) plants which forms a ring-shape dodecameric particle with a central cavity. The oligomeric form of **SP1** is an exceptionally stable structure that is resistant to proteases (e.g., trypsin, V8, and...

...mass spectrometry (MALDI-TOF-MS), and transmission electron microscopy were used to further characterize the **SP1** dodecamer. Introduction of a single cysteine at the N-terminus of **SP1** enabled the formation of disulfide bridges within the **SP1** dodecamer, concurrent with increased melting point. A six-histidine tag was introduced at the N-terminus of **SP1** to generate 6HSP1, and the Delta NSP1 mutant was generated by a deletion of amino...

...terminus. Both 6HSP1 and ANSP1 maintained their ability to assemble a stable dodecamer. Remarkably, these **SP1** homo-dodecamers were able to re-assemble into stable hetero-dodecamers following co-electro-elution from SDS-PAGE. The exceptional stability of the **SP1** -nano ring and its ability to self-assemble hetero-complexes paves the way to further...
DESCRIPTORS:
ORGANISMS: Populus tremula { **aspen** } (Salicaceae)
CHEMICALS & BIOCHEMICALS: ...stable protein 1 { **SP1** }

1/9,K/2 (Item 2 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)

18206926 BIOSIS NO.: 200500113104
The structural basis of the thermostability of SP1 **, a novel plant (Populus tremula) boiling stable protein**
AUTHOR: Dgany Or; Gonzalez Ana; Sofer Oshrat; Wang Xia; Zolotnitsky Gennady ; Wolf Amnon; Shoham Yuval; Altman Arie; Wolf Sharon G; Shoseyov Oded; Almog Orna (Reprint)
AUTHOR ADDRESS: Fac Hlth SciDept Clin Biochem, Ben Gurion Univ Negev, IL-84105, Beer Sheva, Israel**Israel
AUTHOR E-MAIL ADDRESS: almogo@bgu.ac.il
JOURNAL: Journal of Biological Chemistry 279 (49): p51516-51523 December 3, 2004 2004
MEDIUM: print
ISSN: 0021-9258
DOCUMENT TYPE: Article

RECORD TYPE: Abstract
LANGUAGE: English

ABSTRACT: We previously reported on a new boiling stable protein isolated from **aspen** plants (Populus tremula), which we named **SP1** . **SP1** is a stress-related protein with no significant sequence homology to other stress-related proteins. It is a 108-amino-acid hydrophilic polypeptide with a molecular mass of 12.4 kDa (Wang, W. X., Pelah, D., Alergand, T., Shoseyov, O., and Altman, A. (2002) Plant Physiol. 130, 865 - 875) and is found in an oligomeric form. Preliminary electron microscopy studies and matrix-assisted laser desorption ionization time-of-flight mass spectrometry experiments showed that **SP1** is a dodecamer composed of two stacking hexamers. We performed a SDS-PAGE analysis, a differential scanning calorimetric study, and crystal structure determination to further characterize **SP1** . SDS-PAGE indicated a spontaneous assembly of **SP1** to one stable oligomeric form, a dodecamer. Differential scanning calorimetric showed that **SP1** has high thermostability i.e. Tm of 107 degreeC (at pH 7.8). The crystal structure of **SP1** was initially determined to 2.4 ANG resolution by multi-wavelength anomalous dispersion method from a crystal belonging to the space group I422. The phases were extended to 1.8 ANG resolution using data from a different crystal form (P21). The final refined molecule includes 106 of the 108 residues and 132 water molecules (on average for each chain). The R-free is 20.1%. The crystal structure indicated that the **SP1** molecule has a ferredoxin-like fold. Strong interactions between each two molecules create a stable dimer. Six dimers associate to form a ring-like-shaped dodecamer strongly resembling the particle visualized in the electron microscopy studies. No structural similarity was found between the crystal structure of **SP1** and the crystal structure of other stress-related proteins such as small heat shock proteins, whose structure has been already determined. This structural study further supports our previous report that **SP1** may represent a new family of stress-related proteins with high thermostability and oligomerization.

DESCRIPTORS:
MAJOR CONCEPTS: Biochemistry and Molecular Biophysics; Forestry; Methods and Techniques
BIOSYSTEMATIC NAMES: Salicaceae--Dicotyledones, Angiospermae, Spermatophyta, Plantae
ORGANISMS: Populus tremula { **aspen** } (Salicaceae)--ornamental crop
COMMON TAXONOMIC TERMS: Angiosperms; Dicots; Plants; Spermatophytes; Vascular Plants
CHEMICALS & BIOCHEMICALS: **SP1** ; dimer; ferredoxin; small heat shock protein
METHODS & EQUIPMENT: differential scanning calorimetry--laboratory techniques; electron microscopy--imaging and microscopy techniques, laboratory techniques; matrix-assisted laser desorption/ionization time-of-flight mass spectrometry--laboratory techniques, spectrum analysis techniques; sodium dodecyl sulfate-polyacrylamide gel electrophoresis {SDS-PAGE}--electrophoretic techniques, laboratory techniques
MISCELLANEOUS TERMS: oligomerization; thermostability
CONCEPT CODES:
10060 Biochemistry studies - General
51522 Plant physiology - Chemical constituents
53010 Horticulture - Flowers and ornamentals
53500 Forestry and forest products

BIOSYSTEMATIC CODES:
26695 Salicaceae

The structural basis of the thermostability of SP1 , a novel plant (Populus tremula) boiling stable protein

ABSTRACT: We previously reported on a new boiling stable protein isolated from **aspen** plants (Populus tremula), which we named **SP1** . **SP1** is a stress-related protein with no significant sequence homology to other stress-related proteins...

...studies and matrix-assisted laser desorption ionization time-of-flight mass spectrometry experiments showed that **SP1** is a dodecamer composed of two stacking hexamers. We performed a SDS-PAGE analysis, a differential scanning calorimetric study, and crystal structure determination to further characterize **SP1** . SDS-PAGE indicated a spontaneous assembly of **SP1** to one stable oligomeric form, a dodecamer. Differential scanning calorimetric showed that **SP1** has high thermostability i.e. Tm of 107 degreeC (at pH 7.8). The crystal structure of **SP1** was initially determined to 2.4 ANG resolution by multi-wavelength anomalous dispersion method from...

...for each chain). The R-free is 20.1%. The crystal structure indicated that the **SP1** molecule has a ferredoxin-like fold. Strong interactions between each two molecules create a stable...

...in the electron microscopy studies. No structural similarity was found between the crystal structure of **SP1** and the crystal structure of other stress-related proteins such as small heat shock proteins, whose structure has been already determined. This structural study further supports our previous report that **SP1** may represent a new family of stress-related proteins with high thermostability and oligomerization.
DESCRIPTORS:
ORGANISMS: Populus tremula { **aspen** } (Salicaceae...
CHEMICALS & BIOCHEMICALS: **SP1** ;

1/9,K/3 (Item 3 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)

17823981 BIOSIS NO.: 200400214738
Structural features in the model of a thermostable and stress-resistant protein, SP1 **from** aspen .
AUTHOR: Rathore Ravindranath S (Reprint); Narasimhamurthy T
AUTHOR ADDRESS: Department of Physics, Indian Institute of Science, Bangalore, 560 012, India**India
AUTHOR E-MAIL ADDRESS: newdrugdesign@yahoo.com
JOURNAL: Journal of Biomolecular Structure and Dynamics 21 (5): p651-655 April 2004 2004
MEDIUM: print
ISSN: 0739-1102 _(ISSN print)

DOCUMENT TYPE: Article
RECORD TYPE: Abstract
LANGUAGE: English

ABSTRACT: A three dimensional theoretical model of **SP1** (stable protein 1), which is resistant to high temperature and biotic-stresses, is presented here. The model was generated by the application of homology modeling technique. The conformational rigidity imparted to the fold by the presence of hydrogen-bonded, C5, C7, C10 and C13 structures in the loop regions, multiple aromatic - aromatic interactions at the protein interior and on the surface, in addition to salt-links and hydrogen-bonds are primarily the major factors, responsible for the increased stability of protein. The putative protein family is characterized by motifs, E-x(0,1)-L-x-(AEGQS) and V-x(2,3)-L-x-(ADEGST) and the active site in the tertiary structure is formed by conserved aromatic and isoleucine clusters.

DESCRIPTORS:
MAJOR CONCEPTS: Biochemistry and Molecular Biophysics; Models and Simulations--Computational Biology
BIOSYSTEMATIC NAMES: Salicaceae--Dicotyledones, Angiospermae, Spermatophyta, Plantae
ORGANISMS: Populus tremula { **aspen** } (Salicaceae)
COMMON TAXONOMIC TERMS: Angiosperms; Dicots; Plants; Spermatophytes; Vascular Plants
CHEMICALS & BIOCHEMICALS: stable protein 1 { **SP1** }--stress-resistance, thermostability
METHODS & EQUIPMENT: homology modeling--mathematical and computer techniques
MISCELLANEOUS TERMS: three dimensional theoretical model--structural features
CONCEPT CODES:
04500 Mathematical biology and statistical methods
10060 Biochemistry studies - General
10515 Biophysics - Biocybernetics
51522 Plant physiology - Chemical constituents
BIOSYSTEMATIC CODES:
26695 Salicaceae

Structural features in the model of a thermostable and stress-resistant protein, SP1 **from** aspen **.**

ABSTRACT: A three dimensional theoretical model of **SP1** (stable protein 1), which is resistant to high temperature and biotic-stresses, is presented here...
DESCRIPTORS:
ORGANISMS: Populus tremula { **aspen** } (Salicaceae)
CHEMICALS & BIOCHEMICALS: stable protein 1 { **SP1** }--

1/9,K/4 (Item 4 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)

17403150 BIOSIS NO.: 200300361869
A novel chaperone to stabilize Horseradish Peroxidase (HRP) based conjugates in ELISA products: An alternate plant protein to Bovine Serum Albumin (BSA).
AUTHOR: Mehra M (Reprint); DeAnda A (Reprint); Draviam E (Reprint); Ponni Y ; Wolf A; Wang W; Shosegov O; Altman A
AUTHOR ADDRESS: Biotecx, Houston, TX, USA**USA

JOURNAL: Clinical Chemistry 49 (S6): pA117 June 2003 2003
MEDIUM: print
CONFERENCE/MEETING: 55th Annual Meeting of the AACC (American Association for Clinical Chemistry) Philadelphia, PA, USA July 20-24, 2003; 20030720
SPONSOR: American Association for Clinical Chemistry
ISSN: 0009-9147
DOCUMENT TYPE: Meeting; Meeting Abstract
RECORD TYPE: Citation
LANGUAGE: English
REGISTRY NUMBERS: 67-64-1: acetone; 50-01-1: guanidine hydrochloride; 9003-99-0: horseradish peroxidase; 39450-01-6: proteinase K; 9002-07-7: trypsin
ENZYME COMMISSION NUMBER: EC 1.11.1.7: horseradish peroxidase; EC 3.4.21.64 : proteinase K; EC 3.4.21.4: trypsin
DESCRIPTORS:
MAJOR CONCEPTS: Biochemistry and Molecular Biophysics
BIOSYSTEMATIC NAMES: Enterobacteriaceae--Facultatively Anaerobic Gram-Negative Rods, Eubacteria, Bacteria, Microorganisms; Salicaceae--Dicotyledones, Angiospermae, Spermatophyta, Plantae
ORGANISMS: Escherichia coli (Enterobacteriaceae)--expression system; Populus tremula { **aspen** tree} (Salicaceae)
COMMON TAXONOMIC TERMS: Bacteria; Eubacteria; Microorganisms; Angiosperms ; Dicots; Plants; Spermatophytes; Vascular Plants
CHEMICALS & BIOCHEMICALS: SDS; **SP1** --chaperone-like protein; acetone; bovine serum albumin {BSA}; guanidine hydrochloride; horseradish peroxidase--conjugates; phosphate-buffered saline; protein; proteinase K; trypsin
METHODS & EQUIPMENT: Biotecx OptiCoat enzyme immunoassay kit {Biotecx OptiCoat EIA kit}--laboratory kit; ELISA--immunologic techniques, laboratory techniques; Toxoplasma IgG ELISA kit {Toxoplasma immunoglobulin G ELISA kit}--laboratory kit; enzyme immunoassay {EIA}--immunologic techniques, laboratory techniques
CONCEPT CODES:
00520 General biology - Symposia, transactions and proceedings
10060 Biochemistry studies - General
10064 Biochemistry studies - Proteins, peptides and amino acids
10802 Enzymes - General and comparative studies: coenzymes
31000 Physiology and biochemistry of bacteria
51522 Plant physiology - Chemical constituents
BIOSYSTEMATIC CODES:
06702 Enterobacteriaceae
26695 Salicaceae

DESCRIPTORS:
...ORGANISMS: Populus tremula { **aspen** tree} (Salicaceae)
CHEMICALS & BIOCHEMICALS: ... **SP1** --

1/9,K/5 (Item 5 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)

16998828 BIOSIS NO.: 200200592339
Characterization of SP1 **, a stress-responsive, boiling-soluble, homo-oligomeric protein from** aspen
AUTHOR: Wang Wang-Xia; Pelah Dan; Alergand Tal; Shoseyov Oded; Altman Arie (Reprint)

AUTHOR ADDRESS: The Robert H. Smith Institute of Plant Sciences and Genetics in Agriculture and the Otto Warburg Center for Agricultural Biotechnology, Faculty of Agricultural, Food and Environmental Quality Sciences, The Hebrew University of Jerusalem, P.O. Box 12, Rehovot, 76100, Israel**Israel
JOURNAL: Plant Physiology (Rockville) 130 (2): p865-875 October, 2002 2002
MEDIUM: print
ISSN: 0032-0889
DOCUMENT TYPE: Article
RECORD TYPE: Abstract
LANGUAGE: English

ABSTRACT: **sp1** cDNA was isolated from **aspen** (Populus tremula) plants by immunoscreening an expression library using polyclonal antibodies against BspA protein. BspA, which is a boiling-stable protein, accumulates in **aspen** plants in response to water stress and abscisic acid application (Pelah et al., 1995). The **sp1** cDNA was found to encode a 12.4-kD generally hydrophilic protein with a hydrophobic C terminus, which is different from the BspA protein and was termed **SP1** (stable protein 1). Northern-blot analysis revealed that **sp1** encodes a small mRNA (about 0.6 kb) that is expressed in **aspen** plants under non-stress conditions and is accumulated after salt, cold, heat, and desiccation stress, and during the recovery from stress. The **SP1** detected in plants remained soluble upon boiling, migrated both as a 12.4-kD band and a much higher mass of 116 kD on a 17% (w/v) Tricine-sodium dodecyl sulfate-polyacrylamide gel. Comparative protease digestion patterns, amino acid analyses, and the N-terminal sequences of the 12.4- and 116-kD proteins revealed that **SP1** is homo-oligomeric. Furthermore, gel filtration chromatography analysis indicated that **SP1** exists in **aspen** plants as a complex, composed of 12 subunits of 12.4 kD. A large number of sequences deduced from expressed sequence tags and genomic sequences of other organisms with unknown function show high homology to **SP1** . Thus, **SP1** may represent a new protein family. Here, we present the first report on this putative protein family: the cloning, isolation, and characterization of **SP1** , a stress-responsive, boiling-soluble, oligomeric protein.

DESCRIPTORS:
MAJOR CONCEPTS: Biochemistry and Molecular Biophysics
BIOSYSTEMATIC NAMES: Salicaceae--Dicotyledones, Angiospermae, Spermatophyta, Plantae
ORGANISMS: Populus tremula { **aspen** } (Salicaceae)
COMMON TAXONOMIC TERMS: Angiosperms; Dicots; Plants; Spermatophytes; Vascular Plants
CHEMICALS & BIOCHEMICALS: **SP1** --characterization, stress-responsive, boiling soluble, homo-oligomeric protein; **sp1** complementary DNA
METHODS & EQUIPMENT: Northern-blot analysis--analytical method; gel filtration chromatography--analytical method; immunoscreening--immunologic method
MISCELLANEOUS TERMS: cold stress; desiccation stress; heat stress; non-stress conditions; protein family characterization; salt stress; stress recovery
CONCEPT CODES:
10060 Biochemistry studies - General
51522 Plant physiology - Chemical constituents
BIOSYSTEMATIC CODES:
26695 Salicaceae

Characterization of SP1 **, a stress-responsive, boiling-soluble, homo-oligomeric protein from** aspen

ABSTRACT: **sp1** cDNA was isolated from **aspen** (Populus tremula) plants by immunoscreening an expression library using polyclonal antibodies against BspA protein. BspA, which is a boiling-stable protein, accumulates in **aspen** plants in response to water stress and abscisic acid application (Pelah et al., 1995). The **sp1** cDNA was found to encode a 12.4-kD generally hydrophilic protein with a hydrophobic C terminus, which is different from the BspA protein and was termed **SP1** (stable protein 1). Northern-blot analysis revealed that **sp1** encodes a small mRNA (about 0.6 kb) that is expressed in **aspen** plants under non-stress conditions and is accumulated after salt, cold, heat, and desiccation stress, and during the recovery from stress. The **SP1** detected in plants remained soluble upon boiling, migrated both as a 12.4-kD band...

...and the N-terminal sequences of the 12.4- and 116-kD proteins revealed that **SP1** is homo-oligomeric. Furthermore, gel filtration chromatography analysis indicated that **SP1** exists in **aspen** plants as a complex, composed of 12 subunits of 12.4 kD. A large number...

...sequence tags and genomic sequences of other organisms with unknown function show high homology to **SP1** . Thus, **SP1** may represent a new protein family. Here, we present the first report on this putative protein family: the cloning, isolation, and characterization of **SP1** , a stress-responsive, boiling-soluble, oligomeric protein.
DESCRIPTORS:
ORGANISMS: Populus tremula { **aspen** } (Salicaceae)
CHEMICALS & BIOCHEMICALS: **SP1** --...

... **sp1** complementary DNA

1/9,K/6 (Item 1 from file: 24)
DIALOG(R)File 24:CSA Life Sciences Abstracts

0002382470 IP ACCESSION NO: 5476332
Characterization of SP1 **, a Stress-Responsive, Boiling-Soluble, Homo-Oligomeric Protein from** Aspen

Wang, W; Pelah, D; Alergand, T; Shoseyov, O; Altman, A*
The Robert H. Smith Institute of Plant Sciences and Genetics in Agriculture and the Otto Warburg Center for Agricultural Biotechnology, Faculty of Agricultural, Food and Environmental Quality Sciences, The Hebrew University of Jerusalem, P.O. Box 12, Rehovot 76100, Israel, [mailto:altman@agri.huji.ac.il]

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ABSTRACT:
sp1 cDNA was isolated from **aspen** (Populus tremula) plants by immunoscreening an expression library using polyclonal antibodies against BspA protein. BspA, which is a boiling-stable protein, accumulates in **aspen** plants in response to water stress and abscisic acid application (Pelah et al., 1995). The **sp1** cDNA was found to encode a 12.4-kD generally hydrophilic protein with a hydrophobic C terminus, which is different from the BspA protein and was termed **SP1** (stable protein 1). Northern-blot analysis revealed that **sp1** encodes a small mRNA (about 0.6 kb) that is expressed in **aspen** plants under non-stress conditions and is accumulated after salt, cold, heat, and desiccation stress, and during the recovery from stress. The **SP1** detected in plants remained soluble upon boiling, migrated both as a 12.4-kD band and a much higher mass of 116 kD on a 17% (w/v) Tricine-sodium dodecyl sulfate-polyacrylamide gel. Comparative protease digestion patterns, amino acid analyses, and the N-terminal sequences of the 12.4- and 116-kD proteins revealed that **SP1** is homo-oligomeric. Furthermore, gel filtration chromatography analysis indicated that **SP1** exists in **aspen** plants as a complex, composed of 12 subunits of 12.4 kD. A large number of sequences deduced from expressed sequence tags and genomic sequences of other organisms with unknown function show high homology to **SP1** . Thus, **SP1** may represent a new protein family. Here, we present the first report on this putative protein family: the cloning, isolation, and characterization of **SP1** , a stress-responsive, boiling-soluble, oligomeric protein.

DESCRIPTORS: Protein structure; Water stress; Abscisic acid; **sp1** gene; BspA gene; Expressed sequence tags; Populus tremula
SUBJ CATG: 07352, Dicotyledons (miscellaneous)

Characterization of SP1 **, a Stress-Responsive, Boiling-Soluble, Homo-Oligomeric Protein from** Aspen

ABSTRACT:
sp1 cDNA was isolated from **aspen** (Populus tremula) plants by immunoscreening an expression library using polyclonal antibodies against BspA protein. BspA, which is a boiling-stable protein, accumulates in **aspen** plants in response to water stress and abscisic acid application (Pelah et al., 1995). The **sp1** cDNA was found to encode a 12.4-kD generally hydrophilic protein with a hydrophobic C terminus, which is different from the BspA protein and was termed **SP1** (stable protein 1). Northern-blot analysis revealed that **sp1** encodes a small mRNA (about 0.6 kb) that is expressed in **aspen** plants under non-stress conditions and is accumulated after salt, cold, heat, and desiccation stress, and during the recovery from stress. The **SP1** detected in plants remained soluble upon boiling, migrated both as a 12.4-kD band...

...and the N-terminal sequences of the 12.4- and 116-kD proteins revealed that **SP1** is homo-oligomeric. Furthermore, gel filtration chromatography analysis indicated that **SP1** exists in **aspen** plants as a complex, composed of 12 subunits of 12.4 kD. A large number...

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DESCRIPTORS: Protein structure; Water stress; Abscisic acid; **sp1** gene; BspA gene; Expressed sequence tags; Populus tremula

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DIALOG(R)File 34:SciSearch(R) Cited Ref Sci

15482548 Genuine Article#: 076HG Number of References: 21
Title: Aspen SP1 , **an exceptional thermal, protease and detergent-resistant self-assembled nano-particle**
Author(s): Wang WX; Dgany O; Wolf SG; Levy I; Algom R; Pouny Y; Wolf A; Marton I; Altman A; Shoseyov O (REPRINT)
Corporate Source: Hebrew Univ Jerusalem,Fac Agr Food & Environm Qual Sci, Robert H Smith Inst Plant Sci & Genet Agricu,POB 12/IL-76100 Rehovot//Israel/ (REPRINT); Hebrew Univ Jerusalem,Fac Agr Food & Environm Qual Sci, Robert H Smith Inst Plant Sci & Genet Agricu,IL-76100 Rehovot//Israel/; Hebrew Univ Jerusalem,Otto Warburg Ctr Agricultural Biotechnol,IL-76100 Rehovot//Israel/; Fulcrum SP Ltd,IL-46104 Herzliya Pituach//Israel/; Weizmann Inst Sci,Electron Microscopy Unit,IL-76100 Rehovot//Israel/(shoseyov@agri.huji.ac.il)
Journal: BIOTECHNOLOGY AND BIOENGINEERING, 2006, V95, N1 (SEP 5), P161-168
ISSN: 0006-3592 Publication date: 20060905
Publisher: JOHN WILEY & SONS INC, 111 RIVER ST, HOBOKEN, NJ 07030 USA
Language: English Document Type: ARTICLE
Geographic Location: Israel
Journal Subject Category: BIOTECHNOLOGY & APPLIED MICROBIOLOGY
Abstract: Stable protein 1 (**SP1**) is a homo-oligomeric protein isolated from **aspen** (Populus tremula **aspen**) plants which forms a ring-shape dodecameric particle with a central cavity. The oligomeric form of **SP1** is an exceptionally stable structure that is resistant to proteases (e.g., trypsin, V8, and proteinase K), high temperatures, organic solvents, and high levels of ionic detergent. Analytical ultra-centrifugation, chemical cross-linking, matrix-assisted laser-desorption time-of-flight mass spectrometry (MALDI-TOF-MS), and transmission electron microscopy were used to further characterize the **SP1** dodecamer. Introduction of a single cysteine at the N-terminus of **SP1** enabled the formation of disulfide bridges within the **SP1** dodecamer, concurrent with increased melting point. A six-histidine tag was introduced at the N-terminus of **SP1** to generate 6HSP1, and the Delta NSP1 mutant was generated by a deletion of amino acids 2-6 at the N-terminus. Both 6HSP1 and ANSP1 maintained their ability to assemble a stable dodecamer. Remarkably, these **SP1** homo-dodecamers were able to re-assemble into stable hetero-dodecamers following co-electro-elution from SDS-PAGE. The exceptional stability of the **SP1** -nano ring and its ability to self-assemble hetero-complexes paves the way to further research in utilizing this unique protein in nano-biotechnology. (c) 2006 Wiley Periodicals, Inc.
Descriptors--Author Keywords: tremula ; oligomer ; stable ; self-assembly ; nanoparticle
Identifiers--KeyWord Plus(R): ELECTRON-MICROSCOPY; ORGANIC-SOLVENTS; PROTEINS; STABILITY; STRESS
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Title: Aspen SP1 **, an exceptional thermal, protease and detergent-resistant self-assembled nano-particle**

Abstract: Stable protein 1 (**SP1**) is a homo-oligomeric protein isolated from **aspen** (Populus tremula **aspen**) plants which forms a ring-shape dodecameric particle with a central cavity. The oligomeric form of **SP1** is an exceptionally stable structure that is resistant to proteases (e.g., trypsin, V8, and...

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1/9,K/8 (Item 2 from file: 34)

13361789 Genuine Article#: 874MZ Number of References: 33

Title: The structural basis of the thermostability of SP1 **, a novel plant (Populus tremula) boiling stable protein**

Author(s): Dgany O; Gonzalez A; Sofer O; Wang WX; Zolotnitsky G; Wolf A; Shoham Y; Altman A; Wolf SG; Shoseyov O; Almog O (REPRINT)
Corporate Source: Ben Gurion Univ Negev,Fac Hlth Sci, Dept Clin Biochem,IL-84105 Beer Sheva//Israel/ (REPRINT); Ben Gurion Univ Negev,Fac Hlth Sci, Dept Clin Biochem,IL-84105 Beer Sheva//Israel/; Hebrew Univ Jerusalem,Robert H Smith Inst Plant Sci & Genet Agr,IL-76100 Rehovot//Israel/; Stanford Synchrotron Radiat Lab,Menlo Pk//CA/94025; Technion Israel Inst Technol,Dept Food Engn & Biotechnol,

Inst Catalysis Sci & Technol,IL-32000 Haifa//Israel/; Fulcrum SP Ltd,IL-46104 Herzliyya//Israel/; Weizmann Inst Sci,Elect Microscopy Unit,IL-76100 Rehovot//Israel/(almogo@bgu.ac.il)
Journal: JOURNAL OF BIOLOGICAL CHEMISTRY, 2004, V279, N49 (DEC 3), P 51516-51523
ISSN: 0021-9258 Publication date: 20041203
Publisher: AMER SOC BIOCHEMISTRY MOLECULAR BIOLOGY INC, 9650 ROCKVILLE PIKE, BETHESDA, MD 20814-3996 USA
Language: English Document Type: ARTICLE
Geographic Location: Israel; USA
Journal Subject Category: BIOCHEMISTRY & MOLECULAR BIOLOGY
Abstract: We previously reported on a new boiling stable protein isolated from **aspen** plants (Populus tremula), which we named **SP1** . **SP1** is a stress-related protein with no significant sequence homology to other stress-related proteins. It is a 108-amino-acid hydrophilic polypeptide with a molecular mass of 12.4 kDa (Wang, W. X., Pelah, D., Alergand, T., Shoseyov, O., and Altman, A. (2002) Plant Physiol. 130, 865 - 875) and is found in an oligomeric form. Preliminary electron microscopy studies and matrix-assisted laser desorption ionization time-of-flight mass spectrometry experiments showed that **SP1** is a dodecamer composed of two stacking hexamers. We performed a SDS-PAGE analysis, a differential scanning calorimetric study, and crystal structure determination to further characterize **SP1** . SDS-PAGE indicated a spontaneous assembly of **SP1** to one stable oligomeric form, a dodecamer. Differential scanning calorimetric showed that **SP1** has high thermostability i.e. T-m of 107 degreesC (at pH 7.8). The crystal structure of **SP1** was initially determined to 2.4 Angstrom resolution by multi-wavelength anomalous dispersion method from a crystal belonging to the space group I422. The phases were extended to 1.8 Angstrom resolution using data from a different crystal form (P21). The final refined molecule includes 106 of the 108 residues and 132 water molecules (on average for each chain). The R-free is 20.1%. The crystal structure indicated that the **SP1** molecule has a ferredoxin-like fold. Strong interactions between each two molecules create a stable dimer. Six dimers associate to form a ring-like-shaped dodecamer strongly resembling the particle visualized in the electron microscopy studies. No structural similarity was found between the crystal structure of **SP1** and the crystal structure of other stress-related proteins such as small heat shock proteins, whose structure has been already determined. This structural study further supports our previous report that **SP1** may represent a new family of stress-related proteins with high thermostability and oligomerization.
Identifiers--KeyWord Plus(R): HEAT-SHOCK-PROTEIN; WATER-STRESS RESPONSE; CRYSTAL-STRUCTURE; DIFFERENTIAL ACCUMULATION; FREEZING TOLERANCE; SUCROSE SYNTHASE; SOLUBLE SUGARS; REFINEMENT; **ASPEN**; DESICCATION
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WINN MD, 2001, V57, P122, ACTA CRYSTALLOGR D 1
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Title: The structural basis of the thermostability of SP1 **, a novel plant (Populus tremula) boiling stable protein**

Abstract: We previously reported on a new boiling stable protein isolated from **aspen** plants (Populus tremula), which we named **SP1** . **SP1** is a stress-related protein with no significant sequence homology to other stress-related proteins...

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...Identifiers--PROTEIN; WATER-STRESS RESPONSE; CRYSTAL-STRUCTURE; DIFFERENTIAL ACCUMULATION; FREEZING TOLERANCE; SUCROSE SYNTHASE; SOLUBLE SUGARS; REFINEMENT; **ASPEN**; DESICCATION

1/9,K/9 (Item 3 from file: 34)
DIALOG(R)File 34:SciSearch(R) Cited Ref Sci

12596362 Genuine Article#: 802UH Number of References: 32
Title: Structural features in the model of a thermostable and stress-resistant protein, SP1 from aspen
Author(s): Rathore RS (REPRINT) ; Narasimhamurthy T
Corporate Source: Indian Inst Sci,Dept Phys,Bangalore 560012/Karnataka/India/ (REPRINT); Indian Inst Sci,Dept Phys,Bangalore 560012/Karnataka/India/; Indian Inst Sci,Bioinformat Ctr,Bangalore 560012/Karnataka/India/
Journal: JOURNAL OF BIOMOLECULAR STRUCTURE & DYNAMICS, 2004, V21, N5 (APR), P651-655
ISSN: 0739-1102 Publication date: 20040400
Publisher: ADENINE PRESS, 2066 CENTRAL AVE, SCHENECTADY, NY 12304 USA
Language: English Document Type: ARTICLE
Geographic Location: India
Journal Subject Category: BIOCHEMISTRY & MOLECULAR BIOLOGY; BIOPHYSICS
Abstract: A three dimensional theoretical model of **SP1** (stable protein 1), which is resistant to high temperature and biotic-stresses, is presented here. The model was generated by the application of homology modeling technique. The conformational rigidity imparted to the fold by the presence of hydrogen-bonded, C-5, C-7, C-10 and C-13 structures in the loop regions, multiple aromatic-aromatic interactions at the protein interior and on the surface, in addition to salt-links and hydrogen-bonds are primarily the major factors, responsible for the increased stability of protein. The putative protein family is characterized by motifs, E-x(0,1)-L-x-[AEGQS] and V-x(2,3)-L-x-[ADEGST] and the active site in the tertiary structure is formed by conserved aromatic and isoleucine clusters.
Descriptors--Author Keywords: thermostability ; thermophilic ; **SP1** ; aromatic clusters ; homology modeling
Identifiers--KeyWord Plus(R): MOLECULAR-DYNAMICS; SIMULATIONS; PREDICTION; ALIGNMENT; STABILITY; SEQUENCE; DATABASE
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WANG WX, 2002, V130, P865, PLANT PHYSIOL

Title: Structural features in the model of a thermostable and stress-resistant protein, SP1 **from** aspen

Abstract: A three dimensional theoretical model of **SP1** (stable protein 1), which is resistant to high temperature and biotic-stresses, is presented here...

1/9,K/10 (Item 4 from file: 34)

DIALOG(R)File 34:SciSearch(R) Cited Ref Sci

11668924 Genuine Article#: 680LM Number of References: 27

Title: From plant tissue culture to biotechnology: Scientific revolutions, abiotic stress tolerance, and forestry

Author(s): Altman A (REPRINT)

Corporate Source: Hebrew Univ Jerusalem,Robert H Smith Inst Plant Sci & Genet Agr,POB 12/IL-76100 Rehovot//Israel/ (REPRINT); Hebrew Univ Jerusalem,Robert H Smith Inst Plant Sci & Genet Agr,IL-76100 Rehovot//Israel/

Journal: IN VITRO CELLULAR & DEVELOPMENTAL BIOLOGY-PLANT, 2003, V39, N2 (MAR-APR), P75-84

ISSN: 1054-5476 Publication date: 20030300

Publisher: C A B I PUBLISHING, C/O PUBLISHING DIVISION, WALLINGFORD OX10 8DE, OXON, ENGLAND

Language: English Document Type: ARTICLE

Geographic Location: Israel

Journal Subject Category: PLANT SCIENCES; CELL BIOLOGY; DEVELOPMENTAL BIOLOGY

Abstract: Plant biotechnology - especially in vitro regeneration and cell biology, DNA manipulation and biochemical engineering - is already changing the agricultural scene in three major areas: control of plant growth, protecting plants against biotic stress, and production of specialty foods, biochemicals and pharmaceuticals. Plant biotechnology faces several major challenges in the coming decades: alleviating the hazards of abiotic stress (especially salinity, drought, and extreme temperatures), improving pest control, maintenance and improvement of the environment, improvement of food quality and design of 'specialty food' using biochemical engineering, and production of biomaterials. Two parallel research approaches will most likely exist simultaneously in the near future: the transgenic approach (expression of unique genes and specific promoters and transcription factors), and the non-transgenic approach (genomics-assisted gene discovery, marker-assisted selection, efficient mutations, and clonal agriculture). Drought and salinity are the most serious threats to agriculture and to the environment in many parts of the world. Key molecular factors that are being used for genetic engineering of stress-tolerant plants include: over-expression of specific transcription factors, characterization of dehydrin proteins,

over-production of osmoprotectants, expression of water channel proteins and ion transporters, expression and characterization of molecular chaperones, including a novel boiling-stable homo-oligomeric **SP1** protein. Although molecular breeding is routine in agriculture, forest-tree species have been left far behind. However, the increasing demand for wood and its products and the reduction of available harvestable forests has recently led to the introduction of several molecular and biotechnological tools into forest-tree research and improvement. Among these are in vitro propagation, the identification of molecular markers, and genetic engineering for specific traits. Achievements today in plant biotechnology have already surpassed all previous expectations. The full realization and impact of the new developments depend not only on continued successful and innovative research and development activities, but also on a favorable regulatory climate and public acceptance. Plant scientists now have a central role in society.

Descriptors--Author Keywords: abiotic stress ; agricultural biotechnology ; biosafety ; boiling-stable proteins ; chaperones ; compatible solutes ; dehydrins ; drought ; food supply ; forest trees ; gene flow ; ion transporters ; lignin ; osmotic stress ; regeneration ; salinity ; transcription factors ; transformation ; transgenic crops

Identifiers--KeyWord Plus(R): **ASPEN** POPULUS-TREMULA; SIGNALING PATHWAYS; SALT TOLERANCE; CROSS-TALK; PROTEIN; OVEREXPRESSION; TRANSFORMATION; REGENERATION; ARABIDOPSIS

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WANG WX, 2002, BOILING DETERGENT ST
WANG WX, 2001, V560, P285, ACTA HORTIC
ZHU JK, 2001, V6, P66, TRENDS PLANT SCI

...Abstract: ion transporters, expression and characterization of molecular chaperones, including a novel boiling-stable homo-oligomeric **SP1** protein. Although molecular breeding is routine in agriculture, forest-tree species have been left far...

...Identifiers-- **ASPEN** POPULUS-TREMULA; SIGNALING PATHWAYS; SALT TOLERANCE; CROSS-TALK; PROTEIN; OVEREXPRESSION; TRANSFORMATION; REGENERATION; ARABIDOPSIS

1/9,K/11 (Item 5 from file: 34)

DIALOG(R)File 34:SciSearch(R) Cited Ref Sci

11461918 Genuine Article#: 656LT Number of References: 9

Title: Crystallization and preliminary X-ray crystallographic analysis of SP1 , a novel chaperone-like protein

Author(s): Wang WX; Dgany O; Dym O; Altman A; Shoseyov O; Almog O (REPRINT)

Corporate Source: Ben Gurion Univ Negev,Fac Hlth Sci, Dept Clin Biochem,IL-84105 Beer Sheva//Israel/ (REPRINT); Ben Gurion Univ Negev,Fac Hlth Sci, Dept Clin Biochem,IL-84105 Beer Sheva//Israel/; Hebrew Univ Jerusalem,Inst Plant Sci & Genet Agr, Fac Agr Food & Environm Qual Sci,IL-76100 Rehovot//Israel/; Hebrew Univ Jerusalem,Inst Life Sci, Wolfson Ctr Struct Biol,IL-91904 Jerusalem//Israel/

Journal: ACTA CRYSTALLOGRAPHICA SECTION D-BIOLOGICAL CRYSTALLOGRAPHY, 2003 , V59, 3 (MAR), P512-514

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Publisher: BLACKWELL MUNKSGAARD, 35 NORRE SOGADE, PO BOX 2148, DK-1016 COPENHAGEN, DENMARK

Language: English Document Type: ARTICLE

Geographic Location: Israel

Journal Subject Category: BIOCHEMICAL RESEARCH METHODS; BIOCHEMISTRY & MOLECULAR BIOLOGY; BIOPHYSICS; CRYSTALLOGRAPHY

Abstract: **SP1** (108 amino acids) is a boiling-stable stress-responsive protein. It has no significant sequence homology to other stress-related proteins or to small heat-shock proteins (sHsps). **SP1** activity is ATP-independent, similar to other small heat-shock proteins. Based on these features, it is expected that the structure-function relationship of **SP1** will be unique. In this work, the crystallization and preliminary crystallographic data of native **SP1** and its selenomethionine derivative are described. Recombinant **SP1** and its selenomethionine derivative were expressed in Escherichia coli and used for crystallization experiments. **SP1** crystals were grown from 0.1 M HEPES pH 7.5, 20% PEG 3K, 0.2 M NaCl. One to four single crystals appeared in each droplet within a few days and grew to dimensions of about 0.5 x 0.5 x 0.8 mm after about two weeks. Diffraction studies of these crystals at low temperature indicated that they belong to space group I422, with unit-cell parameters a = 89, b = 89, c = 187 Angstrom. Efforts to crystallize the selenomethionine derivative of **SP1** are in progress.

Identifiers--KeyWord Plus(R): WATER-STRESS RESPONSE; **ASPEN** POPULUS-TREMULA; HEAT-SHOCK-PROTEIN; DIFFERENTIAL ACCUMULATION; CRYSTAL-STRUCTURE; SUCROSE SYNTHASE; SOLUBLE SUGARS

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WANG WX, 2002, V130, P865, PLANT PHYSIOL

Title: Crystallization and preliminary X-ray crystallographic analysis of SP1 **, a novel chaperone-like protein**

Abstract: **SP1** (108 amino acids) is a boiling-stable stress-responsive protein. It has no significant sequence homology to other stress-related proteins or to small heat-shock proteins (sHsps). **SP1** activity is ATP-independent, similar to other small heat-shock proteins. Based on these features, it is expected that the structure-function relationship of **SP1** will be unique. In this work, the crystallization and preliminary crystallographic data of native **SP1** and its selenomethionine derivative are described. Recombinant **SP1** and its selenomethionine derivative were expressed in Escherichia coli and used for crystallization experiments. **SP1** crystals were grown from 0.1 M HEPES pH 7.5, 20% PEG 3K, 0...

...parameters a = 89, b = 89, c = 187 Angstrom. Efforts to crystallize the selenomethionine derivative of **SP1** are in progress.

...Identifiers--WATER-STRESS RESPONSE; **ASPEN** POPULUS-TREMULA; HEAT-SHOCK-PROTEIN; DIFFERENTIAL ACCUMULATION; CRYSTAL-STRUCTURE; SUCROSE SYNTHASE; SOLUBLE SUGARS

1/9,K/12 (Item 6 from file: 34)

DIALOG(R)File 34:SciSearch(R) Cited Ref Sci

11100145 Genuine Article#: 605BC Number of References: 48

Title: Characterization of SP1 **, a stress-responsive, boiling-soluble, homo-oligomeric protein from** aspen

Author(s): Wang WX; Pelah D; Alergand T; Shoseyov O; Altman A (REPRINT)

Corporate Source: Hebrew Univ Jerusalem,Fac Agr Food & Environm Qual Sci, Robert H Smith Inst Plant Sci & Genet Agr,POB 12/IL-76100 Rehovot//Israel/ (REPRINT); Hebrew Univ Jerusalem,Fac Agr Food & Environm Qual Sci, Robert H Smith Inst Plant Sci & Genet Agr,IL-76100 Rehovot//Israel/; Hebrew Univ Jerusalem,Fac Agr Food & Environm Qual Sci, Otto Warburg Ctr Agr Biotechnol,IL-76100 Rehovot//Israel/

Journal: PLANT PHYSIOLOGY, 2002, V130, N2 (OCT), P865-875

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Publisher: AMER SOC PLANT BIOLOGISTS, 15501 MONONA DRIVE, ROCKVILLE, MD 20855 USA

Language: English Document Type: ARTICLE

Geographic Location: Israel

Journal Subject Category: PLANT SCIENCES

Abstract: **sp1** cDNA was isolated from **aspen** (Populus tremula) plants by immunoscreening an expression library using polyclonal antibodies against BspA protein. BspA, which is a boiling-stable protein, accumulates in **aspen** plants in response to water stress and abscisic acid application (Pelah et al., 1995). The **sp1** cDNA was found to encode a 12.4-kD generally hydrophilic protein with a hydrophobic C terminus, which is different from the BspA protein and was termed **SP1** (stable protein 1). Northern-blot analysis revealed that **sp1** encodes a small mRNA (about 0.6 kb) that is expressed in **aspen** plants under non-stress conditions and is accumulated after salt, cold, heat, and desiccation stress, and during the recovery from stress. The **SP1** detected in plants remained soluble upon boiling, migrated both as a 12.4-kD band and a much higher mass of 116 kD on a 17% (w/v)

Tricine-sodium dodecyl sulfate-polyacrylamide gel. Comparative protease digestion patterns, amino acid analyses, and the N-terminal sequences of the 12.4- and 116-kD proteins revealed that **SP1** is homo-oligomeric. Furthermore, gel filtration chromatography analysis indicated that **SP1** exists in **aspen** plants as a complex, composed of 12 subunits of 12.4 kD. A large number of sequences deduced from expressed sequence tags and genomic sequences of other organisms with unknown function show high homology to **SP1** . Thus, **SP1** may represent a new protein family. Here, we present the first report on this putative protein family: the cloning, isolation, and characterization of **SP1** , a stress-responsive, boiling-soluble, oligomeric protein.

Identifiers--KeyWord Plus(R): HEAT-SHOCK PROTEINS; PLANT CRATEROSTIGMA-PLANTAGINEUM; SODIUM DODECYL-SULFATE; WATER-STRESS; COLD-ACCLIMATION; MOLECULAR CHAPERONES; ENVIRONMENTAL-STRESS; ARABIDOPSIS-THALIANA; GEL-ELECTROPHORESIS; SIGNALING PATHWAYS

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ZHU JK, 2001, V4, P401, CURR OPIN PLANT BIOL
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Title: Characterization of SP1 **, a stress-responsive, boiling-soluble, homo-oligomeric protein from** aspen

Abstract: **sp1** cDNA was isolated from **aspen** (Populus tremula) plants by immunoscreening an expression library using polyclonal antibodies against BspA protein. BspA, which is a boiling-stable protein, accumulates in **aspen** plants in response to water stress and abscisic acid application (Pelah et al., 1995). The **sp1** cDNA was found to encode a 12.4-kD generally hydrophilic protein with a hydrophobic C terminus, which is different from the BspA protein and was termed **SP1** (stable protein 1). Northern-blot analysis revealed that **sp1** encodes a small mRNA (about 0.6 kb) that is expressed in **aspen** plants under non-stress conditions and is accumulated after salt, cold, heat, and desiccation stress, and during the recovery from stress. The **SP1** detected in plants remained soluble upon boiling, migrated both as a 12.4-kD band...

...and the N-terminal sequences of the 12.4- and 116-kD proteins revealed that **SP1** is homo-oligomeric. Furthermore, gel filtration chromatography analysis indicated that **SP1** exists in **aspen** plants as a complex, composed of 12 subunits of 12.4 kD. A large number...

...sequence tags and genomic sequences of other organisms with unknown function show high homology to **SP1** . Thus, **SP1** may represent a new protein family. Here, we present the first report on this putative protein family: the cloning, isolation, and characterization of **SP1** , a stress-responsive, boiling-soluble, oligomeric protein.

1/9,K/13 (Item 1 from file: 35)
DIALOG(R)File 35:Dissertation Abs Online

01562249 ORDER NO: AADNN-14746
BIODEGRADATION OF LIPIDS BY WOOD SAPSTAINING OPHIOSTOMA SPP. (PINUS CONTORTA, LODGEPOLE PINE, TREMBLING ASPEN **, POPULUS TREMULOIDES)**
Author: GAO, YONG
Degree: PH.D.
Year: 1996
Corporate Source/Institution: THE UNIVERSITY OF BRITISH COLUMBIA (CANADA) (2500)
Adviser: COLETTE BREUIL
Source: VOLUME 58/01-B OF DISSERTATION ABSTRACTS INTERNATIONAL. PAGE 10. 217 PAGES
Descriptors: AGRICULTURE, FORESTRY AND WILDLIFE
Descriptor Codes: 0478
ISBN: 0-612-14746-0

This work was carried out to determine the available lipid nutrients in wood and to understand the biodegradation mechanisms of lipids by

sapstaining Ophiostoma species.

The ability of three sapstaining fungal species Ophiostoma piceae 387N, O. ainoae 701A, and O. piliferum 55H to degrade and utilize the major lipids in the sapwood of lodgepole pine and trembling **aspen** (Populus tremuloides Michx.) was investigated. The fungal growth rate in wood was monitored by quantifying ergosterol extracted from colonized wood. After two weeks colonization, the TGs in wood were degraded by 50% to 80%, which resulted in an accumulation of free FAs in the wood.

Lipases (glycerol ester hydrolases, EC 3.1.1.3) are the enzymes responsible for hydrolyzing TGs into glycerol and FAs which are assimilable by fungal cells. Extracellular lipase activity of O. piceae 387N was detected both in colonized wood and in liquid culture. The effect of various factors (carbon sources, nitrogen sources, and medium pH) on the growth and lipase activity of O. piceae 387N were examined in liquid culture. The extracellular lipase secretion was enhanced in the presence of triglycerides. The composition of a medium was optimized for a high extracellular lipase production, which contained 2% olive oil as a carbon source and 0.5% ammonium sulfate and 3% peptone as nitrogen sources with an initial medium pH of 5.0.

A major extracellular lipase was purified from the liquid culture filtrates of O. piceae 387N by hydrophobic interaction chromatography and anion exchange chromatography. This lipase was characterized as a monomer with a molecular weight of 35 kDa, and was glycosylated, containing 10.1% carbohydrates. It was resolved as a single band on SDS-PAGe (sodium dodecyl sulfate-polyacrylamide gel electrophoresis) gels, whereas 3 bands at pI's 4.3, 4.1 and 3.8 were observed on IEF (isoelectric focusing) gels. Lipolytic stain demonstrated that the three bands on IEF gels were lipolytically active. The 3 isoforms were found to have a same N-terminal sequence as D$\ **sp1** $-V$\sp2$-S$\sp3$-V$\sp4$-T$\sp5$-T$\sp6$-T$\sp7$-D$ \sp8$-I$\sp9$-D$\sp{10}$-A$\sp{11}$-L$\sp{12}$-A$\sp{13}$- F$\sp{14}$-F$\sp{15}$-T$\sp{16}$-Q$\sp{17}$-W$\sp{18}$-A$\sp{19}$-G$\sp{20} .$

The purified O. piceae 387N was stable at pH's 4 to 8 and at temperatures below 40$\sp\circ$C. The pH and temperature optima for activity were approximately pH 5.2 and 30$\sp\circ$C, respectively. Enzyme activity was not influence by N-ethylmaleimide, β-mercaptoethanol, and dithiothreitol, was slightly enhanced by Ca$\sp{2+}$ and Mn$\sp{2+},$ and was severely inhibited by Hg$\sp{2+}$ and Fe$\sp{3+},$ diethyl pyrocarbonate, diethyl p-nitrophenyl phosphate, butyric acid, caproic acid, and SDS. The lipase showed high specificity toward substrates with intermediate and long chain FA residues, and belonged to a group of 1(3) positional specific lipases. The rate of hydrolysis of the lipase toward a triglyceride (1,3-dipalmitoyl-2-oleoyl-glycerol) was 25-50 fold higher than that toward the waxes (oleyl esters) and cholesteryl esters. Finally, it was conclusively shown that the purified lipase could effectively release fatty acid residues from the triglycerides isolated from wood.

The data and information obtained in this work have contributed to the understanding of the physiological and biochemical features of sapstaining Ophiostoma species. (Abstract shortened by UMI.)

BIODEGRADATION OF LIPIDS BY WOOD SAPSTAINING OPHIOSTOMA SPP. (PINUS CONTORTA, LODGEPOLE PINE, TREMBLING ASPEN **, POPULUS TREMULOIDES)**

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...active. The 3 isoforms were found to have a same N-terminal sequence as D$\ **sp1** $-V$\sp2$-S$\sp3$-V$\sp4$-T$\sp5$-T$\sp6$-T$\sp7$-D$ \sp8$-I...

1/9,K/14 (Item 1 from file: 50)
DIALOG(R)File 50:CAB Abstracts

0008769009 CAB Accession Number: 20053029347
The structural basis of the thermostability of SP1 **, a novel plant (Populus tremula) boiling stable protein.**
Dgany, O.; Gonzalez, A.; Sofer, O.; Wang WangXia; Zolotnitsky, G.; Wolf, A.; Shoham, Y.; Altman, A.; Wolf, S. G.; Shoseyov, O.; Almog, O.
Author email address: almogo@bgu.ac.il
Department of Clinical Biochemistry, Faculty of Health Sciences, Ben-Gurion University, Beer-Sheva 84105, Israel.
Journal of Biological Chemistry vol. 279 (49): p.51516-51523
Publication Year: 2004
ISSN: 0021-9258
Digital Object Identifier: 10.1074/jbc.M409952200
Publisher: American Society for Biochemistry and Molecular Biology Inc Bethesda, USA
Language: English Record Type: Abstract
Document Type: Journal article
We previously reported on a new boiling stable protein isolated from **aspen** plants (Populus tremula), which we named **SP1** . **SP1** is a stress-related protein with no significant sequence homology to other stress-related proteins. It is a 108-amino-acid hydrophilic polypeptide with a molecular mass of 12.4 kDa (Wang, W. X., Pelah, D., Alergand, T., Shoseyov, O., and Altman, A. (2002) Plant Physiol. 130, 865-875) and is found in an oligomeric form. Preliminary electron microscopy studies and matrix-assisted laser desorption ionization time-of-flight mass spectrometry experiments showed that **SP1** is a dodecamer composed of two stacking hexamers. We performed a SDS-PAGE analysis, a differential scanning calorimetric study, and crystal structure determination to further characterize **SP1** . SDS-PAGE indicated a spontaneous assembly of **SP1** to one stable oligomeric form, a dodecamer. Differential scanning calorimetric showed that **SP1** has high thermostability i.e. T SUB m of 107(deg)C (at pH 7.8). The crystal structure of **SP1** was initially determined to 2.4 A resolution by multi-wavelength anomalous dispersion method from a crystal belonging to the space group I422. The phases were extended to 1.8 A resolution using data from a different crystal form (P21). The final refined molecule includes 106 of the 108 residues and 132 water molecules (on average for each chain). The R -free is 20.1%. The crystal structure indicated that the **SP1** molecule has a ferredoxin-like fold. Strong interactions between each two molecules create a stable dimer. Six dimers associate to form a ring-like-shaped dodecamer strongly resembling the particle visualized in the electron microscopy studies. No structural similarity was found between the crystal structure of **SP1** and the crystal structure of other stress-related proteins such as small heat shock proteins, whose structure has been already determined. This structural study further supports our previous report that **SP1** may represent a new family of stress-related proteins with high thermostability and oligomerization.
33 ref.

DESCRIPTORS: amino acids; chemical composition; plant composition;

polypeptides; proteins; temperature
ORGANISM DESCRIPTORS: Populus tremula
BROADER TERMS: Populus; Salicaceae; Salicales; dicotyledons; angiosperms; Spermatophyta; plants
CABICODES: Plant Composition (FF040); Forests and Forest Trees (Biology and Ecology) (KK100)

The structural basis of the thermostability of SP1 **, a novel plant (Populus tremula) boiling stable protein.**

We previously reported on a new boiling stable protein isolated from **aspen** plants (Populus tremula), which we named **SP1** . **SP1** is a stress-related protein with no significant sequence homology to other stress-related proteins...

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1/9,K/15 (Item 2 from file: 50)
DIALOG(R)File 50:CAB Abstracts

0008327572 CAB Accession Number: 20023173335
Characterization of SP1 **, a stress-responsive, boiling-soluble, homo-oligomeric protein from** aspen .
Wang WangXia; Pelah, D.; Alergand, T.; Shoseyov, O.; Altman, A.
Author email address: altman@agri.huji.ac.il
The Robert H. Smith Institute of Plant Sciences and Genetics in Agriculture, Faculty of Agricultural, Food and Environmental Quality Sciences, The Hebrew University of Jerusalem, P.O. Box 12, Rehovot 76100, Israel.
Plant Physiology vol. 130 (2): p.865-875
Publication Year: 2002
ISSN: 0032-0889
Digital Object Identifier: 10.1104/pp.002436
Publisher: American Society of Plant Biologists Rockville, USA
Language: English Record Type: Abstract
Document Type: Journal article

The **sp1** cDNA was isolated from **aspen** (Populus tremula) plants by immunoscreening an expression library using polyclonal antibodies against BspA protein. BspA, which is a boiling-stable protein, accumulates in **aspen** plants in response to water stress and abscisic acid application. The **sp1** cDNA was found to encode a 12.4-kD generally hydrophilic protein with a hydrophobic C terminus, which is different from the BspA protein and was termed **SP1** (stable protein 1). Northern-blot analysis revealed that **sp1** encodes a small mRNA (about 0.6 kb) that is expressed in **aspen** plants under non-stress conditions and is accumulated after salt, cold, heat, and desiccation stress, and during recovery from stress. The **SP1** detected in plants remained soluble upon boiling, migrated both as a 12.4-kD band and a much higher mass of 116 kD on a 17% (w/v) tricine-sodium dodecyl sulfate-polyacrylamide gel. Comparative protease digestion patterns, amino acid analyses, and the N-terminal sequences of the 12.4- and 116-kD proteins revealed that **SP1** is homo-oligomeric. Furthermore, gel filtration chromatography analysis indicated that **SP1** exists in **aspen** plants as a complex, composed of 12 subunits of 12.4 kD. A large number of sequences deduced from expressed sequence tags and genomic sequences of other organisms with unknown function show high homology to **SP1** . Thus, **SP1** may represent a new protein family. Here, we present the first report on this putative protein family: the cloning, isolation, and characterization of **SP1** , a stress-responsive, boiling-soluble, oligomeric protein.
48 ref.

DESCRIPTORS: amino acid sequences; characterization; cold stress; DNA cloning; DNA sequencing; genome analysis; genomes; heat stress; messenger RNA; molecular weight; nucleotide sequences; plant proteins; solubility; stress; stress response; water stress
ORGANISM DESCRIPTORS: Populus tremula
BROADER TERMS: Populus; Salicaceae; Salicales; dicotyledons; angiosperms; Spermatophyta; plants
CABICODES: Plant Breeding and Genetics (FF020); Plant Physiology and Biochemistry (FF060); Forests and Forest Trees (Biology and Ecology) (KK100); Molecular Biology and Molecular Genetics, (Discontinued March 2000, Reinstated and Revised June 2002) (ZZ360)

Characterization of SP1 **, a stress-responsive, boiling-soluble, homo-oligomeric protein from** aspen .

The **sp1** cDNA was isolated from **aspen** (Populus tremula) plants by immunoscreening an expression library using polyclonal antibodies against BspA protein. BspA, which is a boiling-stable protein, accumulates in **aspen** plants in response to water stress and abscisic acid application. The **sp1** cDNA was found to encode a 12.4-kD generally hydrophilic protein with a hydrophobic C terminus, which is different from the BspA protein and was termed **SP1** (stable protein 1). Northern-blot analysis revealed that **sp1** encodes a small mRNA (about 0.6 kb) that is expressed in **aspen** plants under non-stress conditions and is accumulated after salt, cold, heat, and desiccation stress, and during recovery from stress. The **SP1** detected in plants remained soluble upon boiling, migrated both as a 12.4-kD band...

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1/9,K/16 (Item 1 from file: 71)
DIALOG(R)File 71:ELSEVIER BIOBASE

03477831 2006256662
Aspen SP1 , **an exceptional thermal, protease and detergent-resistant self-assembled nano-particle**
Wang W.-X.; Dgany O.; Wolf S.G.; Levy I.; Algom R.; Pouny Y.; Wolf A.; Marton I.; Altman A.; Shoseyov O.
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Journal: Biotechnology and Bioengineering, 95/1 (161-168), 2006, United States
PUBLICATION DATE: September 5, 2006
CODEN: BIBIA
ISSN: 0006-3592 eISSN: 1097-0290
DOCUMENT TYPE: Article
LANGUAGES: English SUMMARY LANGUAGES: English
NO. OF REFERENCES: 21

Stable protein 1 (**SP1**) is a homo-oligomeric protein isolated from **aspen** (Populus tremula **aspen**) plants which forms a ring-shape dodecameric particle with a central cavity. The oligomeric form of **SP1** is an exceptionally stable structure that is resistant to proteases (e.g., trypsin, V8, and proteinase K), high temperatures, organic solvents, and high levels of ionic detergent. Analytical ultra-centrifugation, chemical cross-linking, matrix-assisted laser-desorption time-of-flight mass spectrometry (MALDI-TOF-MS), and transmission electron microscopy were used to further characterize the **SP1** dodecamer. Introduction of a single cysteine at the N-terminus of **SP1** enabled the formation of disulfide bridges within the **SP1** dodecamer, concurrent with increased melting point. A six-histidine tag was introduced at the N-terminus of **SP1** to generate 6HSP1, and the DELTANSP1 mutant was generated by a deletion of amino acids 2-6 at the N-terminus. Both 6HSP1 and DELTANSP1 maintained their ability to assemble a stable dodecamer. Remarkably, these **SP1** homo-dodecamers were able to re-assemble into stable hetero-dodecamers following co-electro-elution from SDS-PAGE. The exceptional stability of the **SP1** -nano ring and its ability to self-assemble hetero-complexes paves the way to further research in utilizing this unique protein in nano-biotechnology. (c) 2006 Wiley Periodicals, Inc.

DESCRIPTORS:
Tremula; Oligomer; Stable; Self-assembly; Nanoparticle

SPECIES DESCRIPTORS:
Populus tremula

CLASSIFICATION CODE AND DESCRIPTION:
99 - General

Aspen SP1 , **an exceptional thermal, protease and detergent-resistant self-assembled nano-particle**

Stable protein 1 (**SP1**) is a homo-oligomeric protein isolated from **aspen** (Populus tremula **aspen**) plants which forms a ring-shape dodecameric particle with a central cavity. The oligomeric form of **SP1** is an exceptionally stable structure that is resistant to proteases (e.g., trypsin, V8, and...

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...terminus. Both 6HSP1 and DELTANSP1 maintained their ability to assemble a stable dodecamer. Remarkably, these **SP1** homo-dodecamers were able to re-assemble into stable hetero-dodecamers following co-electro-elution from SDS-PAGE. The exceptional stability of the **SP1** -nano ring and its ability to self-assemble hetero-complexes paves the way to further...

1/9,K/17 (Item 2 from file: 71)
DIALOG(R)File 71:ELSEVIER BIOBASE

02846790 2005001524
The structural basis of the thermostability of SP1 , **a novel plant (Populus tremula) boiling stable protein**
Dgany O.; Gonzalez A.; Sofer O.; Wang W.; Zolotnitsky G.; Wolf A.; Shoham Y.; Altman A.; Wolf S.G.; Shoseyov O.; Almog O.
ADDRESS: O. Almog, Department of Clinical Biochemistry, Faculty of Health Sciences, Ben-Gurion University, Beer-Sheva 84105, Israel
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Journal: Journal of Biological Chemistry, 279/49 (51516-51523), 2004, United States
PUBLICATION DATE: December 3, 2004
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ISSN: 0021-9258
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LANGUAGES: English SUMMARY LANGUAGES: English
NO. OF REFERENCES: 33

We previously reported on a new boiling stable protein isolated from **aspen** plants (Populus tremula), which we named **SP1** . **SP1** is a stress-related protein with no significant sequence homology to other stress-related proteins. It is a 108-amino-acid hydrophilic polypeptide with a molecular mass of 12.4 kDa (Wang, W. X., Pelah, D., Alergand, T., Shoseyov, O., and Altman, A. (2002) Plant Physiol. 130, 865-875) and is found in an oligomeric form. Preliminary electron microscopy studies and matrix-assisted laser desorption ionization time-of-fliglit mass spectrometry experiments showed that **SP1** is a dodecamer composed of two stacking hexamers. We performed a SDS-PAGE analysis, a differential scanning calorimetric study, and crystal structure determination to further

characterize **SP1** . SDS-PAGE indicated a spontaneous assembly of **SP1** to one stable oligomeric form, a dodecamer. Differential scanning calorimetric showed that **SP1** has high thermostability i.e. TSUBm of 107degreesC (at pH 7.8). The crystal structure of **SP1** was initially determined to 2.4 A resolution by multi-wave-length anomalous dispersion method from a crystal belonging to the space group 1422. The phases were extended to 1.8 A resolution using data from a different crystal form (P21). The final refined molecule includes 106 of the 108 residues and 132 water molecules (on average for each chain). The R-free is 20.1%. The crystal structure indicated that the **SP1** molecule has a ferredoxin-like fold. Strong interactions between each two molecules create a stable dimer. Six dimers associate to form a ring-like-shaped dodecamer strongly resembling the particle visualized in the electron microscopy studies. No structural similarity was found between the crystal structure of **SP1** and the crystal structure of other stress-related proteins such as small heat shock proteins, whose structure has been already determined. This structural study further supports our previous report that **SP1** may represent a new family of stress-related proteins with high thermostability and oligomerization.

SPECIES DESCRIPTORS:
Populus tremula

CLASSIFICATION CODE AND DESCRIPTION:
82.2.8 - PROTEIN BIOCHEMISTRY / STRUCTURAL STUDIES / Folding, Unfolding and Stability
82.12 - PROTEIN BIOCHEMISTRY / OTHER PROTEINS
92.1.1.6 - PLANT SCIENCE / BIOCHEMISTRY / Molecular Biology / Proteins

The structural basis of the thermostability of SP1 **, a novel plant (Populus tremula) boiling stable protein**

We previously reported on a new boiling stable protein isolated from **aspen** plants (Populus tremula), which we named **SP1** . **SP1** is a stress-related protein with no significant sequence homology to other stress-related proteins...
...studies and matrix-assisted laser desorption ionization time-of-fliglit mass spectrometry experiments showed that **SP1** is a dodecamer composed of two stacking hexamers. We performed a SDS-PAGE analysis, a differential scanning calorimetric study, and crystal structure determination to further characterize **SP1** . SDS-PAGE indicated a spontaneous assembly of **SP1** to one stable oligomeric form, a dodecamer. Differential scanning calorimetric showed that **SP1** has high thermostability i.e. TSUBm of 107degreesC (at pH 7.8). The crystal structure of **SP1** was initially determined to 2.4 A resolution by multi-wave-length anomalous dispersion method...

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1/9,K/18 (Item 3 from file: 71)
DIALOG(R)File 71:ELSEVIER BIOBASE

02597552 2004072167
Structural Features in the Model of a Thermostable and Stress-resistant Protein, SP1 from aspen
Rathore R.S.; Narasimhamurthy T.
ADDRESS: R.S. Rathore, Department of Physics, Indian Institute of Science, Bangalore 560 012, India
EMAIL: newdrugdesign@yahoo.com
Journal: Journal of Biomolecular Structure and Dynamics, 21/5 (651-655), 2004, United States
CODEN: JBSDD
ISSN: 0739-1102
DOCUMENT TYPE: Article
LANGUAGES: English SUMMARY LANGUAGES: English
NO. OF REFERENCES: 33

A three dimensional theoretical model of **SP1** (stable protein 1), which is resistant to high temperature and biotic-stresses, is presented here. The model was generated by the application of homology modeling technique. The conformational rigidity imparted to the fold by the presence of hydrogen-bonded, CSUB5, CSUB7, CSUB10 and CSUB13 structures in the loop regions, multiple aromatic - aromatic interactions at the protein interior and on the surface, in addition to salt-links and hydrogen-bonds are primarily the major factors, responsible for the increased stability of protein. The putative protein family is characterized by motifs, E-x(0,1)-L-x-[AEGQS] and V-x(2,3)-L-x-[ADEGST] and the active site in the tertiary structure is formed by conserved aromatic and isoleucine clusters.

DESCRIPTORS:
Thermostability; Thermophilic; **SP1** ; Aromatic clusters; Homology modeling

CLASSIFICATION CODE AND DESCRIPTION:
82.2.8 - PROTEIN BIOCHEMISTRY / STRUCTURAL STUDIES / Folding, Unfolding and Stability

MOLECULAR SEQUENCE DATABANK NUMBER:
GENBANK/S30515/(REFERRED NUMBER)
GENBANK/NP 566569/(REFERRED NUMBER)
GENBANK/NP 568422/(REFERRED NUMBER)
GENBANK/NP 866824/(REFERRED NUMBER)
GENBANK/ZP 00058996/(REFERRED NUMBER)

Structural Features in the Model of a Thermostable and Stress-resistant Protein, SP1 from aspen

A three dimensional theoretical model of **SP1** (stable protein 1), which is resistant to high temperature and biotic-stresses, is presented here...

DESCRIPTORS:
Thermostability; Thermophilic; **SP1** ; Aromatic clusters; Homology modeling

1/9,K/19 (Item 4 from file: 71)

02161342 2002247956

Characterization of SP1 , a stress-responsive, boiling-soluble, homo-oligomeric protein from aspen

Wang W.-X.; Pelah D.; Alergand T.; Shoseyov O.; Altman A.
ADDRESS: A. Altman, Robert H. Smith Inst. of Plant Sci., Otto Warburg Ctr. Agric. Biotech., Faculty of Agricultural, P.O. Box 12, Rehovot 76100, Israel
EMAIL: altman@agri.huji.ac.il

Journal: Plant Physiology, 130/2 (865-875), 2002, United States
CODEN: PLPHA
ISSN: 0032-0889
DOCUMENT TYPE: Article
LANGUAGES: English SUMMARY LANGUAGES: English
NO. OF REFERENCES: 48

sp1 cDNA was isolated from **aspen** (Populus tremula) plants by immunoscreening an expression library using polyclonal antibodies against BspA protein. BspA, which is a boiling-stable protein, accumulates in **aspen** plants in response to water stress and abscisic acid application (Pelah et al., 1995). The **sp1** cDNA was found to encode a 12.4-kD generally hydrophilic protein with a hydrophobic C terminus, which is different from the BspA protein and was termed **SP1** (stable protein 1). Northern-blot analysis revealed that **sp1** encodes a small mRNA (about 0.6 kb) that is expressed in **aspen** plants under non-stress conditions and is accumulated after salt, cold, heat, and desiccation stress, and during the recovery from stress. The **SP1** detected in plants remained soluble upon boiling, migrated both as a 12.4-kD band and a much higher mass of 116 kD on a 17% (w/v) Tricine-sodium dodecyl sulfate-polyacrylamide gel. Comparative protease digestion patterns, amino acid analyses, and the N-terminal sequences of the 12.4- and 116-kD proteins revealed that **SP1** is homo-oligomeric. Furthermore, gel filtration chromatography analysis indicated that **SP1** exists in **aspen** plants as a complex, composed of 12 subunits of 12.4 kD. A large number of sequences deduced from expressed sequence tags and genomic sequences of other organisms with unknown function show high homology to **SP1** . Thus, **SP1** may represent a new protein family. Here, we present the first report on this putative protein family: the cloning, isolation, and characterization of **SP1** , a stress-responsive, boiling-soluble, oligomeric protein.

SPECIES DESCRIPTORS:
Populus tremula

CLASSIFICATION CODE AND DESCRIPTION:
92.1.1.6 - PLANT SCIENCE / BIOCHEMISTRY / Molecular Biology / Proteins
92.5.1 - PLANT SCIENCE / STRESS PHYSIOLOGY / Stress
92.1.1.1 - PLANT SCIENCE / BIOCHEMISTRY / Molecular Biology / Gene structure, regulation and function
84.5.26 - GENETICS AND MOLECULAR BIOLOGY / EUKARYOTIC GENETICS / Higher Plant Genetics
84.5.16 - GENETICS AND MOLECULAR BIOLOGY / EUKARYOTIC GENETICS / Biochemical Genetics
84.5.1 - GENETICS AND MOLECULAR BIOLOGY / EUKARYOTIC GENETICS / Extrachromosomal DNA

MOLECULAR SEQUENCE DATABANK NUMBER:
EMBL/AJ276517/(SUBMITTED NUMBER)

Characterization of SP1 **, a stress-responsive, boiling-soluble, homo-oligomeric protein from** aspen

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...and the N-terminal sequences of the 12.4- and 116-kD proteins revealed that **SP1** is homo-oligomeric. Furthermore, gel filtration chromatography analysis indicated that **SP1** exists in **aspen** plants as a complex, composed of 12 subunits of 12.4 kD. A large number...

...sequence tags and genomic sequences of other organisms with unknown function show high homology to **SP1** . Thus, **SP1** may represent a new protein family. Here, we present the first report on this putative protein family: the cloning, isolation, and characterization of **SP1** , a stress-responsive, boiling-soluble, oligomeric protein.

1/9,K/20 (Item 1 from file: 73)
DIALOG(R)File 73:EMBASE

14019737 EMBASE No: 2006434283
Aspen SP1 **, an exceptional thermal, protease and detergent-resistant self-assembled nano-particle**
Wang W.-X.; Dgany O.; Wolf S.G.; Levy I.; Algom R.; Pouny Y.; Wolf A.; Marton I.; Altman A.; Shoseyov O.
O. Shoseyov, Department of Biological Chemistry, Weizmann Institute of Science, Rehovot 76100 Israel
AUTHOR EMAIL: shoseyov@agri.huji.ac.il
Biotechnology and Bioengineering (BIOTECHNOL. BIOENG.) (United States)
05 SEP 2006, 95/1 (161-168)
CODEN: BIBIA ISSN: 0006-3592 eISSN: 1097-0290
DOCUMENT TYPE: Journal ; Article
LANGUAGE: ENGLISH SUMMARY LANGUAGE: ENGLISH
NUMBER OF REFERENCES: 21

Stable protein 1 (**SP1**) is a homo-oligomeric protein isolated from **aspen** (Populus tremula **aspen**) plants which forms a ring-shape dodecameric particle with a central cavity. The oligomeric form of **SP1** is an exceptionally stable structure that is resistant to proteases (e.g., trypsin, V8, and proteinase K), high temperatures, organic solvents, and high levels of ionic detergent. Analytical ultra-centrifugation, chemical cross-linking, matrix-assisted laser-desorption time-of-flight mass

spectrometry (MALDI-TOF-MS), and transmission electron microscopy were used to further characterize the **SP1** dodecamer. Introduction of a single cysteine at the N-terminus of **SP1** enabled the formation of disulfide bridges within the **SP1** dodecamer, concurrent with increased melting point. A six-histidine tag was introduced at the N-terminus of **SP1** to generate 6HSP1, and the DELTANSP1 mutant was generated by a deletion of amino acids 2-6 at the N-terminus. Both 6HSP1 and DELTANSP1 maintained their ability to assemble a stable dodecamer. Remarkably, these **SP1** homo-dodecamers were able to re-assemble into stable hetero-dodecamers following co-electro-elution from SDS-PAGE. The exceptional stability of the **SP1** -nano ring and its ability to self-assemble hetero-complexes paves the way to further research in utilizing this unique protein in nano-biotechnology. (c) 2006 Wiley Periodicals, Inc.

DRUG DESCRIPTORS:
*pregnancy specific beta1 glycoprotein
cysteine; histidine
MEDICAL DESCRIPTORS:
nanoparticle; **aspen** ; oligomerization; temperature; centrifugation; cross linking; matrix assisted laser desorption ionization time of flight mass spectrometry; transmission electron microscopy; amino terminal sequence; polyacrylamide gel electrophoresis; Escherichia coli; nanotechnology; nonhuman; animal cell; article
CAS REGISTRY NO.: 4371-52-2, 52-89-1, 52-90-4 (cysteine); 645-35-2, 7006-35-1, 71-00-1 (histidine)
SECTION HEADINGS:
027 Biophysics, Bioengineering and Medical Instrumentation
029 Clinical and Experimental Biochemistry

Aspen SP1 , **an exceptional thermal, protease and detergent-resistant self-assembled nano-particle**

Stable protein 1 (**SP1**) is a homo-oligomeric protein isolated from **aspen** (Populus tremula **aspen**) plants which forms a ring-shape dodecameric particle with a central cavity. The oligomeric form of **SP1** is an exceptionally stable structure that is resistant to proteases (e.g., trypsin, V8, and...

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MEDICAL DESCRIPTORS:
nanoparticle; **aspen** ; oligomerization; temperature; centrifugation; cross linking; matrix assisted laser desorption ionization time of flight mass spectrometry...

1/9,K/21 (Item 2 from file: 73)
DIALOG(R)File 73:EMBASE

12943483 EMBASE No: 2005003494
The structural basis of the thermostability of SP1 **, a novel plant (Populus tremula) boiling stable protein**
Dgany O.; Gonzalez A.; Sofer O.; Wang W.; Zolotnitsky G.; Wolf A.; Shoham Y.; Altman A.; Wolf S.G.; Shoseyov O.; Almog O.
O. Almog, Department of Clinical Biochemistry, Faculty of Health Sciences, Ben-Gurion University, Beer-Sheva 84105 Israel
AUTHOR EMAIL: almogo@bgu.ac.il
Journal of Biological Chemistry (J. BIOL. CHEM.) (United States) 03 DEC 2004, 279/49 (51516-51523)
CODEN: JBCHA ISSN: 0021-9258
DOCUMENT TYPE: Journal ; Article
LANGUAGE: ENGLISH SUMMARY LANGUAGE: ENGLISH
NUMBER OF REFERENCES: 33

We previously reported on a new boiling stable protein isolated from **aspen** plants (Populus tremula), which we named **SP1** . **SP1** is a stress-related protein with no significant sequence homology to other stress-related proteins. It is a 108-amino-acid hydrophilic polypeptide with a molecular mass of 12.4 kDa (Wang, W. X., Pelah, D., Alergand, T., Shoseyov, O., and Altman, A. (2002) Plant Physiol. 130, 865-875) and is found in an oligomeric form. Preliminary electron microscopy studies and matrix-assisted laser desorption ionization time-of-fliglit mass spectrometry experiments showed that **SP1** is a dodecamer composed of two stacking hexamers. We performed a SDS-PAGE analysis, a differential scanning calorimetric study, and crystal structure determination to further characterize **SP1** . SDS-PAGE indicated a spontaneous assembly of **SP1** to one stable oligomeric form, a dodecamer. Differential scanning calorimetric showed that **SP1** has high thermostability i.e. TSUBm of 107degreesC (at pH 7.8). The crystal structure of **SP1** was initially determined to 2.4 A resolution by multi-wave-length anomalous dispersion method from a crystal belonging to the space group 1422. The phases were extended to 1.8 A resolution using data from a different crystal form (P21). The final refined molecule includes 106 of the 108 residues and 132 water molecules (on average for each chain). The R-free is 20.1%. The crystal structure indicated that the **SP1** molecule has a ferredoxin-like fold. Strong interactions between each two molecules create a stable dimer. Six dimers associate to form a ring-like-shaped dodecamer strongly resembling the particle visualized in the electron microscopy studies. No structural similarity was found between the crystal structure of **SP1** and the crystal structure of other stress-related proteins such as small heat shock proteins, whose structure has been already determined. This structural study further supports our previous report that **SP1** may represent a new family of stress-related proteins with high thermostability and oligomerization.

DRUG DESCRIPTORS:
*heat shock protein--endogenous compound--ec
ferredoxin; dimer; unclassified drug
MEDICAL DESCRIPTORS:
*thermostability; *protein structure
aspen ; molecular weight; electron microscopy; matrix assisted laser desorption ionization time of flight mass spectrometry; polyacrylamide gel

electrophoresis; crystal structure; differential scanning calorimetry; oligomerization; amino acid sequence; sequence analysis; protein interaction; nonhuman; article; priority journal
DRUG TERMS (UNCONTROLLED): stable protein 1--endogenous compound--ec
CAS REGISTRY NO.: 9040-09-9 (ferredoxin)
SECTION HEADINGS:
029 Clinical and Experimental Biochemistry

The structural basis of the thermostability of SP1 **, a novel plant (Populus tremula) boiling stable protein**

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MEDICAL DESCRIPTORS:
aspen ; molecular weight; electron microscopy; matrix assisted laser desorption ionization time of flight mass spectrometry; polyacrylamide...

1/9,K/22 (Item 3 from file: 73)
DIALOG(R)File 73:EMBASE

12520165 EMBASE No: 2004119421
Structural Features in the Model of a Thermostable and Stress-resistant Protein, SP1 **from** aspen
Rathore R.S.; Narasimhamurthy T.
R.S. Rathore, Department of Physics, Indian Institute of Science, Bangalore 560 012 India
AUTHOR EMAIL: newdrugdesign@yahoo.com
Journal of Biomolecular Structure and Dynamics (J. BIOMOL. STRUCT. DYN.) (United States) 2004, 21/5 (651-655)
CODEN: JBSDD ISSN: 0739-1102
DOCUMENT TYPE: Journal ; Article
LANGUAGE: ENGLISH SUMMARY LANGUAGE: ENGLISH

NUMBER OF REFERENCES: 33

A three dimensional theoretical model of **SP1** (stable protein 1), which is resistant to high temperature and biotic-stresses, is presented here. The model was generated by the application of homology modeling technique. The conformational rigidity imparted to the fold by the presence of hydrogen-bonded, CSUB5, CSUB7, CSUB10 and CSUB13 structures in the loop regions, multiple aromatic - aromatic interactions at the protein interior and on the surface, in addition to salt-links and hydrogen-bonds are primarily the major factors, responsible for the increased stability of protein. The putative protein family is characterized by motifs, E-x(0,1)-L-x-[AEGQS] and V-x(2,3)-L-x-[ADEGST] and the active site in the tertiary structure is formed by conserved aromatic and isoleucine clusters.
;
MOLECULAR SEQUENCE NUMBER: GENBANK, S30515; GENBANK, NP 566569; GENBANK, NP 568422; GENBANK, NP 866824; GENBANK, ZP 00058996
DRUG DESCRIPTORS:
*protein
aromatic amino acid; isoleucine; unclassified drug
MEDICAL DESCRIPTORS:
*protein structure; *thermostability; * **aspen**
theoretical model; stress; sequence homology; protein conformation; protein folding; hydrogen bond; protein cross linking; protein interaction; protein family; protein motif; protein tertiary structure; nonhuman; article; nucleotide sequence; priority journal
DRUG TERMS (UNCONTROLLED): stable protein 1
CAS REGISTRY NO.: 67254-75-5 (protein); 7004-09-3, 73-32-5 (isoleucine)
SECTION HEADINGS:
029 Clinical and Experimental Biochemistry

Structural Features in the Model of a Thermostable and Stress-resistant Protein, SP1 **from** aspen

A three dimensional theoretical model of **SP1** (stable protein 1), which is resistant to high temperature and biotic-stresses, is presented here...
MEDICAL DESCRIPTORS:
*protein structure; *thermostability; * **aspen**

1/9,K/23 (Item 1 from file: 143)
DIALOG(R)File 143:Biol. & Agric. Index

1962559 H.W. WILSON RECORD NUMBER: BBAI06145228
Aspen SP1 , **An Exceptional Thermal, Protease and Detergent-Resistant Self-Assembled Nano-Particle**
Wang, Wang-Xia
Dgany, Or; Wolf, Sharon Grayer
Biotechnology & Bioengineering v. 95 no1 (September 5 2006) p. 161-8
ISSN: 0006-3592 LANGUAGE: English
RECORD STATUS: Corrected or revised record

DESCRIPTORS: Molecular self-assembly; Nanoparticles; Plant proteins--Biotechnology

Aspen SP1 , **An Exceptional Thermal, Protease and Detergent-Resistant Self-Assembled Nano-Particle**

1/9,K/24 (Item 2 from file: 143)
DIALOG(R)File 143:Biol. & Agric. Index

1813106 H.W. WILSON RECORD NUMBER: BBAI05101903
The Structural Basis of the Thermostability of SP1 **, a Novel Plant (Populus tremula) Boiling Stable Protein**
Dgany, Or
Gonzalez, Ana; Sofer, Oshrat
Journal of Biological Chemistry v. 279 no49 (December 3 2004) p. 51516-23
ISSN: 0021-9258 LANGUAGE: English
RECORD STATUS: New record

DESCRIPTORS: European **aspen** ; Plant proteins--Crystallography; Heat shock proteins--Plants

The Structural Basis of the Thermostability of SP1 **, a Novel Plant (Populus tremula) Boiling Stable Protein**

DESCRIPTORS: European **aspen** ;

1/9,K/25 (Item 3 from file: 143)
DIALOG(R)File 143:Biol. & Agric. Index

1575262 H.W. WILSON RECORD NUMBER: BBAI02112845
Characterization of SP1 **, a Stress-Responsive, Boiling-Soluble, Homo-Oligomeric Protein from** Aspen
Wang, Wang-Xia
Pelah, Dan; Alergand, Tal
Plant Physiology v. 130 no2 (Oct. 2002) p. 865-75
DOCUMENT TYPE: Feature Article ISSN: 0032-0889 LANGUAGE: English
RECORD STATUS: New record

DESCRIPTORS: Plant proteins; European **aspen**

Characterization of SP1 **, a Stress-Responsive, Boiling-Soluble, Homo-Oligomeric Protein from** Aspen

...DESCRIPTORS: European **aspen**

1/9,K/26 (Item 1 from file: 144)
DIALOG(R)File 144:Pascal

17965751 PASCAL No.: 07-0026165
Aspen SP1 **, an exceptional thermal, protease and detergent-resistant self-assembled nano-particle**
WANY Wang-Xia; DGANY Or; WOLF Sharon Grayer; LEVY Ilan; ALGOM Rachel; POUNY Yehonathan; WOLF Amnon; MARTON Ira; ALTMAN Arie; SHOSEYOV Oded
The Robert H. Smith Institute of Plant Sciences and Genetics in Agriculture, and the Otto Warburg Center for Agricultural Biotechnology, Faculty of Agricultural, Food and Environmental Quality Sciences, The

Hebrew University of Jerusalem, P.O. Box 12, Rehovot 76100, Israel; Fulcrum SP Ltd., P.O.Box 3206, Herzliya Pituach 46104, Israel; Electron Microscopy Unit, Weizmann Institute of Science, Rehovot 76100, Israel
Journal: Biotechnology and bioengineering, 2006, 95 (1) 161-168
ISSN: 0006-3592 CODEN: BIBIAU Availability: INIST-9164;
354000133472330180
No. of Refs.: 21 ref.
Document Type: P (Serial) ; A (Analytic)
Country of Publication: United States
Language: English
Stable protein 1 (**SP1**) is a homo-oligomeric protein isolated from **aspen** (Populus tremula **aspen**) plants which forms a ring-shape dodecameric particle with a central cavity. The oligomeric form of **SP1** is an exceptionally stable structure that is resistant to proteases (e.g., trypsin, V8, and proteinase K), high temperatures, organic solvents, and high levels of ionic detergent. Analytical ultra-centrifugation, chemical cross-linking, matrix-assisted laser-desorption time-of-flight mass spectrometry (MALDI-TOF-MS), and transmission electron microscopy were used to further characterize the **SP1** dodecamer. Introduction of a single cysteine at the N-terminus of **SP1** enabled the formation of disulfide bridges within the **SP1** dodecamer, concurrent with increased melting point. A six-histidine tag was introduced at the N-terminus of **SP1** to generate 6HSP1, and the ANSP1 mutant was generated by a deletion ofamino acids 2-6 at the N-terminus. Both 6HSP1 and ANSP1 maintained their ability to assemble a stable dodecamer. Remarkably, these **SP1** homo-dodecamers were able to re-assemble into stable hetero-dodecamers following co-electro-elution from SDS-PAGE. The exceptional stability of the **SP1** -nano ring and its ability to self-assemble hetero-complexes paves the way to further research in utilizing this unique protein in nano-biotech-nology.

English Descriptors: Peptidases; Detergent; Resistance; Self assembly; Nanoparticle; Oligomer
Broad Descriptors: Hydrolases; Enzyme; Hydrolases; Enzyme; Hydrolases; Enzima
French Descriptors: Peptidases; Detergent; Resistance; Autoassemblage; Nanoparticule; Oligomere

Classification Codes: 002A31; 215

Aspen SP1 , **an exceptional thermal, protease and detergent-resistant self-assembled nano-particle**

Stable protein 1 (**SP1**) is a homo-oligomeric protein isolated from **aspen** (Populus tremula **aspen**) plants which forms a ring-shape dodecameric particle with a central cavity. The oligomeric form of **SP1** is an exceptionally stable structure that is resistant to proteases (e.g., trypsin, V8, and...

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ofamino acids 2...

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1/9,K/27 (Item 2 from file: 144)
DIALOG(R)File 144:Pascal

15862336 PASCAL No.: 02-0582411
Characterization of SP1 **, a stress-responsive, boiling-soluble, homo-oligomeric protein from** aspen
WANG Wang-Xia; PELAH Dan; ALERGAND Tal; SHOSEYOV Oded; ALTMAN Arie
The Robert H. Smith Institute of Plant Sciences and Genetics in Agriculture and the Otto Warburg Center for Agricultural Biotechnology, Faculty of Agricultural, Food and Environmental Quality Sciences, The Hebrew University of Jerusalem, P.O. Box 12, Rehovot 76100, Israel
Journal: Plant physiology : (Bethesda), 2002, 130 (2) 865-875
ISSN: 0032-0889 CODEN: PPHYA5 Availability: INIST-3000; 354000105348900300
No. of Refs.: 48 ref.
Document Type: P (Serial) ; A (Analytic)
Country of Publication: United States
Language: English
sp1 cDNA was isolated from **aspen** (Populus tremula) plants by immunoscreening an expression library using polyclonal antibodies against BspA protein. BspA, which is a boiling-stable protein, accumulates in **aspen** plants in response to water stress and abscisic acid application (Pelah et al., 1995). The **sp1** cDNA was found to encode a 12.4-kD generally hydrophilic protein with a hydrophobic C terminus, which is different from the BspA protein and was termed **SP1** (stable protein 1). Northern-blot analysis revealed that spl encodes a small mRNA (about 0.6 kb) that is expressed in **aspen** plants under non-stress conditions and is accumulated after salt, cold, heat, and desiccation stress, and during the recovery from stress. The **SP1** detected in plants remained soluble upon boiling, migrated both as a 12.4-kD band and a much higher mass of 116 kD on a 17% (w/v) Tricine-sodium dodecyl sulfate-polyacrylamide gel. Comparative protease digestion patterns, amino acid analyses, and the N-terminal sequences of the 12.4- and 116-kD proteins revealed that **SP1** is homo-oligomeric. Furthermore, gel filtration chromatography analysis indicated that **SP1** exists in **aspen** plants as a complex, composed of 12 subunits of 12.4 kD. A large number of sequences deduced from expressed sequence tags and genomic sequences of other organisms with unknown function show high homology to **SP1** . Thus, **SP1** may represent a new protein family. Here, we present the first report on this putative protein family: the cloning, isolation, and characterization of **SP1** , a stress-responsive, boiling-soluble, oligomeric protein.

English Descriptors: Water stress; Characterization; Isolation; Molecular cloning; Gene; Aminoacid sequence; Thermal stability; Nucleotide sequence ; Oligomer; Populus tremula; Abscisic acid; Salinity; Cold; Heat; Desiccation
Broad Descriptors: Salicaceae; Dicotyledones; Angiospermae; Spermatophyta;

Environmental factor; Hardwood forest tree; Sesquiterpenes; Plant growth substance; Salicaceae; Dicotyledones; Angiospermae; Spermatophyta; Facteur milieu; Arbre forestier feuillu; Sesquiterpene; Substance croissance vegetal; Salicaceae; Dicotyledones; Angiospermae; Spermatophyta; Factor medio; Arbol forestal frondoso; Sesquiterpeno; Substancia crecimiento vegetal

French Descriptors: Stress hydrique; Caracterisation; Isolement; Clonage moleculaire; Gene; Sequence aminoacide; Stabilite thermique; Sequence nucleotide; Oligomere; Populus tremula; Abscissique acide; Salinite; Froid; Chaleur; Dessiccation; Gene **SP1**

Classification Codes: 002A10B06; 002A04C02

Characterization of SP1 **, a stress-responsive, boiling-soluble, homo-oligomeric protein from** aspen

sp1 cDNA was isolated from **aspen** (Populus tremula) plants by immunoscreening an expression library using polyclonal antibodies against BspA protein. BspA, which is a boiling-stable protein, accumulates in **aspen** plants in response to water stress and abscisic acid application (Pelah et al., 1995). The **sp1** cDNA was found to encode a 12.4-kD generally hydrophilic protein with a hydrophobic C terminus, which is different from the BspA protein and was termed **SP1** (stable protein 1). Northern-blot analysis revealed that sp1 encodes a small mRNA (about 0.6 kb) that is expressed in **aspen** plants under non-stress conditions and is accumulated after salt, cold, heat, and desiccation stress, and during the recovery from stress. The **SP1** detected in plants remained soluble upon boiling, migrated both as a 12.4-kD band...

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...French Descriptors: aminoacide; Stabilite thermique; Sequence nucleotide ; Oligomere; Populus tremula; Abscissique acide; Salinite; Froid; Chaleur ; Dessiccation; Gene **SP1**

1/9,K/28 (Item 1 from file: 155)
DIALOG(R)File 155:MEDLINE(R)

21762624 PMID: 16732592
Aspen SP1 **, an exceptional thermal, protease and detergent-resistant self-assembled nano-particle.**
Wang Wang-Xia; Dgany Or; Wolf Sharon Grayer; Levy Ilan; Algom Rachel; Pouny Yehonathan; Wolf Amnon; Marton Ira; Altman Arie; Shoseyov Oded
The Robert H. Smith Institute of Plant Sciences and Genetics in

Agriculture, and the Otto Warburg Center for Agricultural Biotechnology, Faculty of Agricultural, Food and Environmental Quality Sciences, The Hebrew University of Jerusalem, Israel.

Biotechnology and bioengineering (United States) Sep 5 2006, 95 (1) p161-8, ISSN 0006-3592--Print Journal Code: 7502021
Publishing Model Print
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: MEDLINE; Completed
Subfile: INDEX MEDICUS

Stable protein 1 (**SP1**) is a homo-oligomeric protein isolated from **aspen** (Populus tremula **aspen**) plants which forms a ring-shape dodecameric particle with a central cavity. The oligomeric form of **SP1** is an exceptionally stable structure that is resistant to proteases (e.g., trypsin, V8, and proteinase K), high temperatures, organic solvents, and high levels of ionic detergent. Analytical ultra-centrifugation, chemical cross-linking, matrix-assisted laser-desorption time-of-flight mass spectrometry (MALDI-TOF-MS), and transmission electron microscopy were used to further characterize the **SP1** dodecamer. Introduction of a single cysteine at the N-terminus of **SP1** enabled the formation of disulfide bridges within the **SP1** dodecamer, concurrent with increased melting point. A six-histidine tag was introduced at the N-terminus of **SP1** to generate 6HSP1, and the DeltaNSP1 mutant was generated by a deletion of amino acids 2-6 at the N-terminus. Both 6HSP1 and DeltaNSP1 maintained their ability to assemble a stable dodecamer. Remarkably, these **SP1** homo-dodecamers were able to re-assemble into stable hetero-dodecamers following co-electro-elution from SDS-PAGE. The exceptional stability of the **SP1** -nano ring and its ability to self-assemble hetero-complexes paves the way to further research in utilizing this unique protein in nano-biotechnology. (c) 2006 Wiley Periodicals, Inc.

Descriptors: *Crystallization--methods--MT; *Detergents--chemistry--CH; *Nanostructures--chemistry--CH; *Nanostructures--ultrastructure--UL; *Plant Proteins--chemistry--CH; *Plant Proteins--ultrastructure--UL; *Populus --enzymology--EN; Dimerization; Enzyme Activation; Enzyme Stability; Multiprotein Complexes--analysis--AN; Multiprotein Complexes--chemistry --CH; Multiprotein Complexes--ultrastructure--UL; Nanostructures--analysis --AN; Plant Proteins--analysis--AN; Protein Binding; Research Support, Non-U.S. Gov't; Temperature
CAS Registry No.: 0 (Detergents); 0 (Multiprotein Complexes); 0 (Plant Proteins)
Record Date Created: 20060828
Record Date Completed: 20061005

Aspen SP1 , **an exceptional thermal, protease and detergent-resistant self-assembled nano-particle.**

Stable protein 1 (**SP1**) is a homo-oligomeric protein isolated from **aspen** (Populus tremula **aspen**) plants which forms a ring-shape dodecameric particle with a central cavity. The oligomeric form of **SP1** is an exceptionally stable structure that is resistant to proteases (e.g., trypsin, V8, and...

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1/9,K/29 (Item 2 from file: 155)
DIALOG(R)File 155:MEDLINE(R)

15188859 PMID: 15371455

The structural basis of the thermostability of SP1 **, a novel plant (Populus tremula) boiling stable protein.**

Dgany Or; Gonzalez Ana; Sofer Oshrat; Wang Wangxia; Zolotnitsky Gennady; Wolf Amnon; Shoham Yuval; Altman Arie; Wolf Sharon G; Shoseyov Oded; Almog Orna

Department of Clinical Biochemistry, Faculty of Health Sciences, Ben-Gurion University, Beer-Sheva 84105, Israel.

Journal of biological chemistry (United States) Dec 3 2004, 279 (49) p51516-23, ISSN 0021-9258--Print Journal Code: 2985121R

Publishing Model Print-Electronic

Document type: Journal Article

Languages: ENGLISH

Main Citation Owner: NLM

Record type: MEDLINE; Completed

Subfile: INDEX MEDICUS

We previously reported on a new boiling stable protein isolated from **aspen** plants (Populus tremula), which we named **SP1** . **SP1** is a stress-related protein with no significant sequence homology to other stress-related proteins. It is a 108-amino-acid hydrophilic polypeptide with a molecular mass of 12.4 kDa (Wang, W. X., Pelah, D., Alergand, T., Shoseyov, O., and Altman, A. (2002) Plant Physiol. 130, 865-875) and is found in an oligomeric form. Preliminary electron microscopy studies and matrix-assisted laser desorption ionization time-of-flight mass spectrometry experiments showed that **SP1** is a dodecamer composed of two stacking hexamers. We performed a SDS-PAGE analysis, a differential scanning calorimetric study, and crystal structure determination to further characterize **SP1** . SDS-PAGE indicated a spontaneous assembly of **SP1** to one stable oligomeric form, a dodecamer. Differential scanning calorimetric showed that **SP1** has high thermostability i.e. Tm of 107 degrees C (at pH 7.8). The crystal structure of **SP1** was initially determined to 2.4 A resolution by multi-wavelength anomalous dispersion method from a crystal belonging to the space group I422. The phases were extended to 1.8 A resolution using data from a different crystal form (P21). The final refined molecule includes 106 of the 108 residues and 132 water molecules (on average for each chain). The R-free is 20.1%. The crystal structure indicated that the **SP1** molecule has a ferredoxin-like fold. Strong interactions between each two molecules create a stable dimer. Six dimers associate to form a ring-like-shaped dodecamer strongly resembling the particle visualized in the electron microscopy studies. No structural similarity was found between the crystal structure of **SP1** and the crystal structure of other stress-related proteins such as small heat shock proteins, whose structure has been already determined. This structural study further supports our previous report that **SP1** may represent a new

family of stress-related proteins with high thermostability and oligomerization.

Descriptors: *Heat-Shock Proteins--chemistry--CH; *Plant Proteins --chemistry--CH; *Populus--metabolism--ME; Amino Acid Sequence; Calorimetry, Differential Scanning; Crystallography, X-Ray; Dimerization; Electrophoresis, Polyacrylamide Gel; Glutamic Acid--chemistry--CH; Microscopy, Electron; Models, Molecular; Molecular Sequence Data; Peptides --chemistry--CH; Protein Conformation; Protein Structure, Secondary; Research Support, Non-U.S. Gov't; Sequence Homology, Amino Acid; Spectrometry, Mass, Matrix-Assisted Laser Desorption-Ionization; Temperature

Molecular Sequence Databank No.: PDB/1SI9; PDB/1TR0

CAS Registry No.: 0 (Heat-Shock Proteins); 0 (Peptides); 0 (Plant Proteins); 56-86-0 (Glutamic Acid)

Record Date Created: 20041125

Record Date Completed: 20050111

Date of Electronic Publication: 20040914

The structural basis of the thermostability of SP1 **, a novel plant (Populus tremula) boiling stable protein.**

We previously reported on a new boiling stable protein isolated from **aspen** plants (Populus tremula), which we named **SP1** . **SP1** is a stress-related protein with no significant sequence homology to other stress-related proteins...

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1/9,K/30 (Item 3 from file: 155)

DIALOG(R)File 155:MEDLINE(R)

14743068 PMID: 14769057

Structural features in the model of a thermostable and stress-resistant protein, SP1 **from** aspen .

Rathore Ravindranath S; Narasimhamurthy T

Department of Physics, Indian Institute of Science, Bangalore 560 012, India. newdrugdesign@yahoo.com

Journal of biomolecular structure & dynamics (United States) Apr 2004, 21 (5) p651-5, ISSN 0739-1102--Print Journal Code: 8404176
Publishing Model Print
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: MEDLINE; Completed
Subfile: INDEX MEDICUS

A three dimensional theoretical model of **SP1** (stable protein 1), which is resistant to high temperature and biotic-stresses, is presented here. The model was generated by the application of homology modeling technique. The conformational rigidity imparted to the fold by the presence of hydrogen-bonded, C5, C7, C10 and C13 structures in the loop regions, multiple aromatic--aromatic interactions at the protein interior and on the surface, in addition to salt-links and hydrogen-bonds are primarily the major factors, responsible for the increased stability of protein. The putative protein family is characterized by motifs, E-x(0,1)-L-x-[AEGQS] and V-x(2,3)-L-x-[ADEGST] and the active site in the tertiary structure is formed by conserved aromatic and isoleucine clusters.

Descriptors: *Populus--chemistry--CH; *Protein Conformation; *Proteins --chemistry--CH; Amino Acid Sequence; Models, Molecular; Molecular Sequence Data
CAS Registry No.: 0 (Proteins)
Record Date Created: 20040210
Record Date Completed: 20041007

Structural features in the model of a thermostable and stress-resistant protein, SP1 **from** aspen .

A three dimensional theoretical model of **SP1** (stable protein 1), which is resistant to high temperature and biotic-stresses, is presented here...

1/9,K/31 (Item 4 from file: 155)
DIALOG(R)File 155:MEDLINE(R)

13963775 PMID: 12376651
Characterization of SP1 **, a stress-responsive, boiling-soluble, homo-oligomeric protein from** aspen .

Wang Wang-Xia; Pelah Dan; Alergand Tal; Shoseyov Oded; Altman Arie
The Robert H. Smith Institute of Plant Sciences and Genetics in Agriculture Faculty of Agricultural, Food and Environmental Quality Sciences, The Hebrew University of Jerusalem, P.O. Box 12, Rehovot 76100, Israel.

Plant physiology (United States) Oct 2002, 130 (2) p865-75, ISSN 0032-0889--Print Journal Code: 0401224
Publishing Model Print
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
Record type: MEDLINE; Completed
Subfile: INDEX MEDICUS

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Descriptors: *Adaptation, Physiological--physiology--PH; *Populus --genetics--GE; Abscisic Acid--pharmacology--PD; Adaptation, Physiological --genetics--GE; Amino Acid Sequence; Base Sequence; Blotting, Northern; Cloning, Molecular; Cycloheximide--pharmacology--PD; DNA, Complementary --chemistry--CH; DNA, Complementary--genetics--GE; Electrophoresis, Gel, Two-Dimensional; Mannitol--pharmacology--PD; Molecular Sequence Data; Peptide Mapping; Plant Proteins--genetics--GE; Plant Proteins--metabolism --ME; Populus--drug effects--DE; Populus--metabolism--ME; Research Support, Non-U.S. Gov't; Sequence Alignment; Sequence Analysis, DNA; Sodium Chloride--pharmacology--PD; Water--pharmacology--PD; Water--physiology--PH

Molecular Sequence Databank No.: GENBANK/AJ276517

CAS Registry No.: 0 (DNA, Complementary); 0 (Plant Proteins); 21293-29-8 (Abscisic Acid); 66-81-9 (Cycloheximide); 69-65-8 (Mannitol); 7647-14-5 (Sodium Chloride); 7732-18-5 (Water)

Record Date Created: 20021011

Record Date Completed: 20030206

Characterization of SP1 **, a stress-responsive, boiling-soluble, homo-oligomeric protein from** aspen .

sp1 cDNA was isolated from **aspen** (Populus tremula) plants by immunoscreening an expression library using polyclonal antibodies against BspA protein. BspA, which is a boiling-stable protein, accumulates in **aspen** plants in response to water stress and abscisic acid application (Pelah et al., 1995). The **sp1** cDNA was found to encode a 12.4-kD generally hydrophilic protein with a hydrophobic C terminus, which is different from the BspA protein and was termed **SP1** (stable protein 1). Northern-blot analysis revealed that **sp1** encodes a small mRNA (about 0.6 kb) that is expressed in **aspen** plants under non-stress conditions and is accumulated after salt, cold, heat, and desiccation stress, and during the recovery from stress. The **SP1** detected in plants remained soluble upon boiling, migrated both as a 12.4-kD band...

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1/9,K/32 (Item 1 from file: 357)

0406927 DBR Accession No.: 2006-20423

Aspen SP1 , **an exceptional thermal, protease and detergent-resistant self-assembled nano-particle - vector-mediated gene transfer and expression in Escherichia coli for recombinant stable protein-1 production for use in nanobiotechnology**

AUTHOR: WANG WX; DGANY O; WOLF SG; LEVY I; ALGOM R; POUNY Y; WOLF A; MARTON I; ALTMAN A; SHOSEYOV O

CORPORATE AFFILIATE: Hebrew Univ Jerusalem Hebrew Univ Jerusalem Fulcrum SP Ltd Weizmann Inst Sci

CORPORATE SOURCE: Shoseyov O, Hebrew Univ Jerusalem, Fac Agr Food and Environm Qual Sci, Robert H Smith Inst Plant Sci and Genet Agriculture, POB 12, IL-76100 Rehovot, Israel

JOURNAL: BIOTECHNOLOGY AND BIOENGINEERING (95, 1, 161-168) 2006

ISSN: 0006-3592

LANGUAGE: English

ABSTRACT: AUTHOR ABSTRACT - Stable protein 1 (**SP1**) is a homo-oligomeric protein isolated from **aspen** (Populus tremula **aspen**) plants which forms a ring-shape dodecameric particle with a central cavity. The oligomeric form of **SP1** is an exceptionally stable structure that is resistant to proteases (e.g., trypsin, V8, and proteinase K), high temperatures, organic solvents, and high levels of ionic detergent. Analytical ultra-centrifugation, chemical cross-linking, matrix-assisted laser-desorption time-of-flight mass spectrometry (MALDI-TOF-MS), and transmission electron microscopy were used to further characterize the **SP1** dodecamer. Introduction of a single cysteine at the N-terminus of **SP1** enabled the formation of disulfide bridges within the **SP1** dodecamer, concurrent with increased melting point. A six-histidine tag was introduced at the N-terminus of **SP1** to generate 6HSP1, and the Delta NSP1 mutant was generated by a deletion of amino acids 2-6 at the N-terminus. Both 6HSP1 and ANSP1 maintained their ability to assemble a stable dodecamer. Remarkably, these **SP1** homo-dodecamers were able to re-assemble into stable hetero-dodecamers following co-electro-elution from SDS-PAGE. The exceptional stability of the **SP1** -nano ring and its ability to self-assemble hetero-complexes paves the way to further research in utilizing this unique protein in nano-biotechnology. (c) 2006 Wiley Periodicals, Inc. (8 pages)

DESCRIPTORS: **aspen** recombinant protease stable protein-1, six-histidine tag stable protein-1 prep., purification, characterization, crystal struct., vector-mediated gene transfer, expression in Escherichia coli, ultracentrifugation, matrix-assisted laser-desorption time-of-flight mass spectrometry, transmission electron microscopy, site-directed mutagenesis, high temp., SDS conc. effect, appl. nanobiotechnology, protein engineering, protein scaffold plant forest tree thermostable enzyme bacterium sedimentation (25, 38)

SECTION: BIOMANUFACTURING and BIOCATALYSIS-Biocatalyst Isolation and Characterization-GENETIC TECHNIQUES and APPLICATIONS-Gene Expression Techniques and Analysis; BIOMANUFACTURING and BIOCATALYSIS-Biocatalyst Application

Aspen SP1 , **an exceptional thermal, protease and detergent-resistant self-assembled nano-particle - vector-mediated gene transfer...**

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DESCRIPTORS: **aspen** recombinant protease stable protein-1, six-histidine tag stable protein-1 prep., purification, characterization, crystal...

1/9,K/33 (Item 2 from file: 357)

DIALOG(R)File 357:Derwent Biotech Res.

0338849 DBR Accession No.: 2004-11141 PATENT

Novel isolated denaturant which is e.g. boiling- or detergent-stable and/or protease resistant protein having chaperone-like activity, useful for strengthening hair, nail or skin and for inducing wound healing - recombinant enzyme protein production for use in disease therapy and plant engineering

AUTHOR: WANG W; PELAH D; ALEGRAND T; SHOSEYOV O; ALTMAN A; POUNY Y; MARTON I; WOLF A

PATENT ASSIGNEE: YISSUM RES DEV CO HEBREW UNIV JERUSALEM; FULCRUM SP LTD 2004

PATENT NUMBER: WO 200422697 PATENT DATE: 20040318 WPI ACCESSION NO.: 2004-248452 (200423)

PRIORITY APPLIC. NO.: US 233409 APPLIC. DATE: 20020904

NATIONAL APPLIC. NO.: WO 2003IL723 APPLIC. DATE: 20030902

LANGUAGE: English

ABSTRACT: DERWENT ABSTRACT: NOVELTY - An isolated denaturant stable (boiling stable or detergent stable) and/or protease resistant protein (I) having chaperone-like activity and horseradish peroxidase (HRP) protection activity, as determined by HRP protection assay, is new. DETAILED DESCRIPTION - INDEPENDENT CLAIMS are also included for: (1) an isolated nucleic acid (II) comprising a first polynucleotide encoding (I), and a second polynucleotide including a promoter sequence operably linked to the first polynucleotide for directing an expression of (I); (2) a nucleic acid construct comprising (II); (3) a cell transformed with (II); (4) an organism transformed with (II); (5) isolating a gene encoding (I) from a biological source, involves screening an expression library with a polynucleotide encoding (I); (6) an antibody (III)

recognizing one or more epitopes of (I); (7) enriching or isolating (M1) denaturant stable (boiling stable or detergent stable) and/or protease resistant protein from a biological source, involves extracting total proteins from the biological source to obtain a protein extract, boiling the protein extract, collecting the soluble proteins and optionally assaying for chaperone-like activity of the soluble proteins and enriching or isolating the stable protein having chaperone-like activity; (8) isolating a gene encoding (I) from a biological source; (9) identifying a nucleic acid potentially encoding (I), involves searching an electronic library containing several nucleic acid and/or amino acid sequences for sequences having a predetermined degree of identity or homology to any one of the nucleic acid sequences (N1) chosen from 5 fully defined sequences of 567, 593, 357, 497, 366 base pairs as given in the specification, or to any one of the amino acid sequences (A1) chosen from 26 fully defined sequences e.g., 108, 98, 98, 98, 84, 98, 98, 109, 47, 98, 98, 93 and 108 amino acids as given in the specification, or their portions or corresponding to at least 15 bases; (10) isolating a nucleic acid potentially encoding (I); (11) detergent-free isolation of a protease-resistant protein having chaperone-like activity from a biological source, involves extracting total proteins from the biological source, to obtain a protein extract, contacting the protein extract with a protease, and isolating a protease-resistant protein, and optionally assaying the protease-resistant protein for chaperone-like activity; (12) a fusion protein (IV) comprising (I) fused to an additional polypeptide; (13) a transgenic plant expressing (I) above a natural amount of (I) in the plant; (14) rendering a plant more tolerant to a biotic or abiotic stress, involves engineering the plant to express (I) above a natural amount of (I) in the plant; (15) rendering a plant more recoverable from a biotic or abiotic stress, involves engineering the plant to express (I) above a natural amount of (I) in the plant; (16) isolating (M2) a boiling stable protein from a biological source, involves carrying out extracting, and boiling steps of (M1), recovering soluble protein fraction, and optionally assaying the protease resistant protein for chaperone-like activity; (17) a pharmaceutical composition (V) comprising (I) as an active ingredient and a carrier; (18) a hetero complex (VI) comprising an oligomer including several of (I), and at least two different molecules being fused to the oligomer; and (19) increasing a specific activity of a pre-isolated (I) as determined in Units of protecting activity per mg protein, involves autoclaving the pre-isolated (I), or treating the pre-isolated (I) with a protease. BIOTECHNOLOGY - Preferred Protein: The isolated denaturant stable (boiling stable or detergent stable) and/or protease resistant protein (I) has chaperone-like activity and horseradish peroxidase (HRP) protection activity, as determined by HRP protection assay, of at least 10 Units/mg protein, where the HRP protection assay comprises mixing (I) at different final protein concentrations at a predetermined volume with 100 microliters of 5 nM HRP present in 40 mM N-(2-hydroxyethyl)piperazine-N'-ethanesulfonic acid (HEPES) buffer at pH 7.5, thus forming a first reaction mixture, and following incubation of the reaction mixture at 25 degreesC for 16 hours, determining HRP remaining enzymatic activity by mixing 5 microliters of the first reaction mixture with 100 microliters of 3,3',5,5'-tetramethylbenzidine , thus forming a second reaction mixture, incubating the second reaction mixture for 10 minutes, stopping a reaction of the second reaction mixture by an addition of 100 microliters of 1 M sulfuric acid and recording colorimetric change in the second reaction mixture at 435

nm, and units are defined as a dilution factor of (I) at a concentration of 1 mg/ml that confers 50% protection of HRP activity in the HRP protection assay. In (I), the HRP protection activity is of 15000, 10000, 8000, 6000, 5500, 5000, 4500, 4000, 3500, 3000, 2500, 2000, 1500, 1000 or at least 500 Units/mg protein. Preferred Nucleic Acid: In (II), the promoter sequence is a eukaryotic constitutive promoter. The promoter is a plant promoter chosen from a constitutive plant promoter, a tissue specific plant promoter and an inducible plant promoter. The constitutive plant promoter is chosen from Cauliflower mosaic virus (CaMV)35S plant promoter, CamV19S plant promoter, figwort mosaic virus (FMV)34S plant promoter, sugarcane bacilliform badnavirus plant promoter, CsVMV plant promoter, Arabidopsis ACT2/ACT8 actin plant promoter, Arabidopsis ubiquitin UBQ1 plant promoter, barley leaf thionin BTH6 plant promoter, and rice actin plant promoter. The tissue specific plant promoter is chosen from bean phaseolin storage protein plant promoter, DLEC plant promoter, PHSbeta plant promoter, zein storage protein plant promoter, conglutin gamma plant promoter from soybean, AT2S1 gene plant promoter, ACT11 actin plant promoter from Arabidopsis, napA plant promoter from Brassica napus and potato patatin gene plant promoter. The inducible plant promoter is chosen from a light-inducible plant promoter derived from the pea rbcS gene, a plant promoter from the alfalfa rbcS gene, DRE, MYC and MYB plant promoters, which are active in drought, INT, INPS, prxEa, Ha hsp17.7G4 and RD21 plant promoters active in high salinity and osmotic stress, and hsr203J and str246C plant promoters active in pathogenic stress. The first polynucleotide has a sequence at least 60% identical with 6 fully defined sequences of 567, 593, 357, 428, 497 or 366 base pairs as given in the specification, as determined using the BestFit software of the Wisconsin sequence analysis package, utilizing the Smith and Waterman algorithm, where gap weight equals 50, length weight equals 3, average match equals 10 and average mismatch equals -9. (I) Has a sequence at least 60% identical to a fully defined sequence of 108 or 112 amino acids as given in the specification, as determined using the BestFit software of the Wisconsin sequence analysis package, utilizing the Smith and Waterman algorithm, where gap creation penalty equals 8 and gap extension penalty equals 2. (I) Is natively an oligomer. The chaperone-like activity includes heat stabilization of proteins. (II) Further comprises a third polynucleotide encoding an additional protein, and being adjacent and in frame to the first polynucleotide, where the first and third polynucleotides encoding, in combination, a fusion protein of the (I) and the additional protein. Preferred Pharmaceutical Composition: (V) Is packaged in a package and identified in print for use in a wound healing application or strengthening and/or grooming hair, nail or skin application. Preferred Method: Isolating a gene encoding (I) from a biological source, comprises: (a) carrying out the steps of extracting, boiling, collecting, assaying, and isolating of (M1), raising antibodies recognizing the stable protein having chaperone-like activity and screening an expression library with the antibodies; or (b) carrying out the steps of (M1), micro-sequencing the stable protein to obtain at least a partial amino acid sequence, designing an oligonucleotide corresponding to the amino acid sequence and screening a library with the oligonucleotide. Isolating a nucleic acid potentially encoding (I) comprises: (a) screening a cDNA or genomic library with a polynucleotide of at least 17 bases and at least 60% identical to a contiguous portion of 6 fully defined sequences of 567, 593, 357, 428, 497 or 366 base pairs as given in the specification; or (b) providing at least one pair of oligonucleotides

each being at least 15 bases in length, including one or more oligonucleotides corresponding to (N1), and selected for amplifying a nucleic acid having a degree of identity with, or encoding proteins exhibiting homology to (A1), contacting at least one pair of oligonucleotides with a sample of nucleic acid and amplifying the nucleic acid having degree of identity with, or encoding proteins exhibiting homology to (A1), and using nucleic acid having degree of identity with, or encoding proteins exhibiting homology to (A1) for isolating a nucleic acid potentially encoding (I). (M2) further involves digesting the protein extract with a protease. Preferred Hetero Complex: In (VI), the two different molecules comprise a first enzyme and a second enzyme, that catalyze sequential or different reactions in a synthesis or degradation pathway. The two different molecules comprise at least a binding molecule and a reporter molecule. Preferred Fusion Protein: (I) Is fused to the additional polypeptide through a peptide bond or a cross-linker, where (IV) has an oligomeric form. ACTIVITY - Vulnerary; Nootropic; Neuroprotective. No biological data given. MECHANISM OF ACTION - Prevents aggregation of aggregating proteins; Inducer of immune response (claimed). USE - (I) Is useful for preventing an aggregating protein from aggregating into an aggregate, which involves causing an effective amount of (I) to become in contact with aggregating protein. (I) Is useful for de-aggregating aggregates of an aggregating protein, which involves causing an effective amount of (I) to become in contact with the aggregate. (I) Is useful for stabilizing a protein against denaturing conditions, which involves causing an effective amount of (I) to become in contact with the protein. (I) Is useful for protecting an enzyme preparation from reduction in enzymatic activity, which involves adding (I) to the enzyme preparation in an amount sufficient for protecting the enzyme preparation from reduction in enzymatic activity. (I) is useful for repairing at least a portion of lost enzymatic activity of an enzyme preparation, which involves adding (I) to the enzyme preparation, in an amount sufficient for repairing portion of the lost enzymatic activity of the enzyme preparation. (I) is useful for increasing cell migration, which involves exposing the cells to (I), in an amount sufficient for increasing cell migration. (I) is useful for accelerating or inducing wound healing, which involves administering (I) on to a wound, in an amount sufficient for accelerating or inducing wound healing. (I) is useful for strengthening or grooming hair, nail or skin, which involves administering (I) onto the hair, nail or skin sufficient for strengthening or grooming the hair, nail or skin. (I) is useful for treating a disease associated with protein aggregation of an aggregating protein, which involves administering (I) to a subject who is in need, in an amount sufficient for de-aggregating and/or preventing aggregation of the aggregating protein such as beta-amyloid or prion. (I) is useful for increasing a binding avidity of a binding molecule, which involves displaying multiple copies of the binding molecule on a surface of an oligomer of (I), where the binding molecule is chosen from a receptor, ligand, enzyme, substrate, inhibitor, antibody or antigen. (I) is useful in administering a polypeptide to an animal having a immune system by reducing an immune response against the polypeptide, which involves administering the polypeptide being fused to (I), to the animal and thus reducing the immune response against the polypeptide as compared to the immune response that is developed by administering the polypeptide alone to the animal. (III) is useful for isolating gene encoding (I), which involves screening an expression library with (III). (IV) is useful in immunization, which

involves subjecting an immune system of a mammal to (IV) (claimed). (I) is useful for treating a disease such as Alzheimer's disease and prion associated diseases e.g., encephalus spongyform, by preventing aggregation of aggregating proteins. ADVANTAGE - (I) retains its activity and oligomerability also when forming a fusion protein. EXAMPLE - Boiling stable protein fractions of **aspen** , tomato M82, VF36 and pine were prepared as follows: Crude plant extracts were centrifuged for 10 minutes and supernatants were transferred to fresh tubes. The supernatants were subjected to a 10-minutes boiling session, then kept on ice for 5 minutes and centrifuged for 10 minutes. Resulting supernatants were precipitated by adding 4 volumes of cold acetone, and centrifuged for 10 minutes. Boiling stable proteins were then recovered by dissolving the pellets in 10 mM Tris-hydrochloric acid buffer (pH 7.5). The total boiling-stable proteins were separated on a 17% sodium dodecyl sulfate (SDS)-tricine polyacrylamide gel electrophoresis (PAGE), during which two bands of 66 and 116 kDa band were obtained. The 66 kDa band was found to represent a germin-like protein. Acetone-precipitated boiling-stable proteins of **aspen** plant were dissolved in 1X tricine-SDS sample buffer (100 mM Tris-hydrochloric acid pH 6.8, 20% glycerol, 1% SDS, 0.025% Coomassie blue), and then separated on a preparative 17% polyacrylamide tricine-SDS gel. Major bands corresponding to stable proteins (SP)-1 (116 kDa oligomer and 12.4 kDa monomer) protein were excised from the gel. **SP1** oligomer and monomer were electro-eluted separately, in a dialysis bag. The eluted product was further dialyzed against 500 volumes of 10 mM Tris-hydrochloric acid overnight at 4degreesC, followed by acetone precipitation and centrifugation. Purified **SP1** was obtained by dissolving the pellet in 10 mM Tris-hydrochloric acid. The prepared stable protein when maintained with horseradish peroxidase, was found to maintain its activity. (176 pages)

DESCRIPTORS: recombinant denaturant stable protease resistant, chaperone-like protein, horseradish peroxidase, prep., vector-mediated gene transfer expression in host cell, antibody, fusion protein, appl. transgenic plant construction, biotic, abiotic stress tolerance, aggregating protein prevention, protein stabilization, enzyme loss act. repair, cell migration, wound healing induction, grooming hair, Alzheimer disease, prion associated disease, encephalus spongyform therapy plant Armoracia rusticana enzyme EC-1.11.1.7 crop improvement vulnerary nootropic neuroprotective DNA sequence protein sequence (23, 22)

SECTION: THERAPEUTICS-Protein Therapeutics-GENETIC TECHNIQUES and APPLICATIONS-Gene Expression Techniques and Analysis; DISEASE-Central Nervous System-AGRICULTURAL BIOTECHNOLOGY-Plant Genetic Engineering; DISEASE-Other Diseases

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